ISSN: 2456-1878

International Journal of Environment Agriculture and Biotechnology

(IJEAB)

An open access Peer Reviewed International Journal

DOI: 10.22161/ijeab.51

Vol.- 5 | Issue - 1 | Jan-Feb 2020

editor@ijeab.com | http://www.ijeab.com/

International Editorial Board/ Reviewer Board

FOREWORD

I am pleased to put into the hands of readers Volume-5; Issue-1: Jan-Feb 2020 of **"International Journal of Environment, Agriculture and Biotechnology (IJEAB) (ISSN: 2456-1878)",** an international journal which publishes peer reviewed quality research papers on a wide variety of topics related to **Environment, Agriculture and Biotechnology**. Looking to the keen interest shown by the authors and readers, the editorial board has decided to release issue with DOI (Digital Object Identifier) from CrossRef also, now using DOI paper of the author is available to the many libraries. This will motivate authors for quick publication of their research papers. Even with these changes our objective remains the same, that is, to encourage young researchers and academicians to think innovatively and share their research findings with others for the betterment of mankind.

I thank all the authors of the research papers for contributing their scholarly articles. Despite many challenges, the entire editorial board has worked tirelessly and helped me to bring out this issue of the journal well in time. They all deserve my heartfelt thanks.

Finally, I hope the readers will make good use of this valuable research material and continue to contribute their research finding for publication in this journal. Constructive comments and suggestions from our readers are welcome for further improvement of the quality and usefulness of the journal.

With warm regards.

Editor-in-Chief
Date: Mar, 2020

Vol-5, Issue-1, Jan-Feb 2020

(*DOI: 10.22161/ijeab.51*)

https://dx.doi.org/10.22161/ijeab.51.1 *ISSN: 2456-1878*

"Salt lick potentials in Ecotourism Management of Borgu Sector": Kainji Lake National Park, Nigeria

Wahab, M.K.A[1*]; Alarape, A. A.[2]; Halidu, S. K.[3]; and Idowu, I. A.[1]

[1]Department of Fisheries and Wildlife Management, Osun State University, Osogbo, Nigeria.
[2]Department of Wildlife and Ecotourism Management, University of Ibadan, Nigeria.
[3]Department of Wildlife and Ecotourism Management, Federal College of Wildlife Management, New- Bussa, Nigeria

Abstract**—** *Ecotourism is a form of tourism undertaken to view and / or encounter wildlife in a range of settings. One of such eco-destinations in which wildlife can be viewed is salt licks. The study was undertaken at Borgu sector of Kainji Lake National Park. Data collection was carried out using systematic random sampling for the selection of salt lick sites, direct and indirect fauna observation using transect lines to determine the level of site utilization. Secondary data from the park management and laboratory analysis of salt lick, soil samples to detect the mineral compositions and richness of the licks as relevant to ecotourism activities in the park was examined. Observation revealed that the iron concentration (a trace element) is high in salt lick 1 (332.33), while the lowest was recorded in salt lick 6 (36.36).It was also revealed that the content of calcium (a major element) is high in salt lick 1 (4.22), with the least recorded in salt lick 6 (0.40). It was perceived that salt lick 3 and 4 were least utilized during the dry seasons.*

It is important to know that the mineral content of salt lick sites can be a factor affecting its utilization by fauna resources. It was observed that, turn-out of tourists visiting the park fluctuates; as a result of anthropogenic activities and other limiting factors.The overall benefits derived from salt licks for wildlife health,majorly through herbivores are crucial in maintaining a healthy wildlife community for their reproduction and survival.

Keywords**— Conservation, Biodiversity Management, Ecotourism potential, Management tool, Eco-destination.**

I. INTRODUCTION

Development of ecotourism management had long been recognized since the period of independence when the National Policies on wildlife management was established in the country. The establishment of protected areas for conservation management basically centers on protection and conservation of sites for National development. In recent times, the National wildlife policy now centers equal emphasis on all aspects of wildlife utilization; recreation, tourism, bush meat production and preservation of gene pool. The dire need to manage wildlife for tourism development was placed first among other goals in many states of the federation, due to immense benefits derivable from tourism; revenue generation, foreign exchange, value addition to the local product and job creation. Tourismoffers about 1 million jobs in California State of America (Odumosu 2003 as cited by Ijeomah 2007) as well as 200,000 jobs in Sri lanka (Ashley, 2005).Tourism isas well identified as one of the key areas by the Nigerian governmentfor the nation's socio-economic transformation (Ijeomah, 2007). The transformation can be maximized throughproper evaluation of tourism performance in eco-destinations as an effective tool on which ecotourism management of a National park can be based.Tourism and conservation objectives are complementary and jointly achievable in National Parks and game reserves, although tourism is supposed to be secondary to conservation as described by the proponents of ecological tourism (Yunis, 2003. On this basis, Nigeria National Parks alongside its major mandate to conserve flora and fauna in various ecological zones are also scheduled to act as catalysts for the development of ecotourism (NTDC, 2005).Wildlife tourism often referred to as ecotourism is a form of tourism undertaken to view and / or encounter wildlife in a range of settings, from capture semi captive, to in the wild and it encompasses a variety of interactions from passive observation to feeding and /or touching the species viewed

https://dx.doi.org/10.22161/ijeab.51.1 *ISSN: 2456-1878*

(Newsome *et al*, 2005).One of such eco-destinations in which wildlife can be viewed is salt licks. Salt licks are key places and locations for the ecological dynamics of wildlife communities in protected areas. It is a natural mineral deposit area where animals visit frequently and actively for mineral uptake through licking or geophagy.Geophagy is a behavioral patterncommon among ruminants for addressing mineral deficiencies, deficiencies in major and trace elements which may aid to ease digestiveproblems or buffer the effects of dietary toxins(Stephenson et al. 2011). The location of a mineral lick in a protected area strongly influence the movement and distribution of ungulate populations (Heimer 1974; Simmons 1982; Watts and Schemnitz, 1985). Unlike forage vegetation patterns, which are non-static and vary with natural disturbance and weather over time, mineral licks are a static resource that may be used by many generations of a population.Species composition of visitors and frequency of visits may differ from one lick to the next (Tobler 2009; Blake 2010), such variation may reflect differences in mineral composition of the soil among different sites (Abrahams et al. 1999) predation may affect the types of species and numbers of visits to different licks (Izawa et al. 1993) or differences among species in habitat preferences and availability of licks in different habitats.Kainji Lake National Park is an area of high biodiversity value and includes a number of saltlick sites frequently used by different herbivores and carnivores. However the ungulate species are prone to flee when exposed to human disturbance(Stankowich 2008). These small, localized areas areimportant resources for all ungulate species; their preservation on the landscape is vitalin conservation management approaches (Dormaar and Walker 1996; Rea et al. 2004).

II. MATERIALS AND METHOD

Kainji Lake National Park is Nigeria's first experiment at establishing and managing a National park. The National park was established in 1979 (under decrees 46 of 29th July, 1979)making it one of the most important National parks in Africa, it is highly endowed with many flora and fauna resources. The park is located between latitudes 9º40'N to 10º30'N and longitudes 4º30'E to 5º50'E. It is made up of two non-contiguous sectors, the Borgu and Zugurma sectors with Borgu sector comprising 3,970.83 km2 (74.3%) and the Zugurma sector covering an area of 1,370 km2 (25.7%). The savanna climate of the park is responsible for the distinct wet and dry seasons. The six main broad vegetationclassificationsin the Park are;

(i) *Burkea Africana /Deuteriummicrocarpus woodland savanna*
(ii) *Diospyrusmespiliformis*dry forest
(iii) Riparian forest and woodland
(iv) *Terminaliamacroptera*tree savanna
(v) *Isoberliniatomentosa*woodland and
(vi) *Isoberliniadoka*, savanna woodland.

The mean temperature observed during the wet season is about 30ºC which drops to 28º C during the dry season associated with the north easternharmattan winds. Rainfall is a major climatic element in the park facilitating vegetation growth and the hydrology of the rivers. The mean annuals rainfall is about 1200mm.

III. RESULTS

Table 1: Mineral analysis of nine soil samples

Sample No	Sample ID	Cu3+ (mg/L)	Mg2+ (mg/L)	Mn2+ (mg/L)	Fe2+ (mg/L)	Zn2+ (mg/L)	Ca2+ (mg/L)
1	Salt lick 1	0.37	7.69	2.24	332.33	1.398	4.22
2	Salt lick 2	0.26	5.58	1.172	257.03	1.20	2.49
3	Salt lick 3	0.15	2.96	0.70	99.92	1.267	0.58
4	Salt lick 4	0.37	5.91	1.17	737.50	0.941	1.28
5	Salt lick 5	0.20	1.95	1.53	155.37	0.74	1.28
6	Salt lick 6	0.09	2.05	N.D	36.36	0.68	0.47
7	Salt lick 7	0.26	5.97	1.054	298.16	0.68	1.56
8	Salt lick 8	0.20	3.46	1.17	148.20	0.68	9.20
9	Salt lick 9	0.37	7.095	0.94	298.16	N.D	1.28

Note: N.D- Not Detected.

https://dx.doi.org/10.22161/ijeab.51.1 *ISSN: 2456-1878*

Reconnaissance survey of the park (Borgu sector) was carried out to familiarize with the park resources and salt licks present which can function as a tool in enhancing ecotourism development in the protected area. Soil samples were collected from nine different salt lick points randomly selected from the total of twenty five salt licks in the Borgu sector of the park. These samples were analyzed in Osun state college of Agriculture's laboratory using Atomic Absorption Spectrophotometer (AAS) Model PG 990 to detect the various mineral compositions. The soil samples were digested using concentrated Nitric acid, 1g weighed into a digestion flask, and 10ml of Acid added. The mixture was heated at 200^0C for one hour to breakdown the elements composition.

Secondary data was gathered to elicit information from the park management on tourist's visitation in the park for the year 2007-2017. These data were subjected to descriptive analysis involving the use of charts, graphs and tables.

Concentration of Major Elements in the Sampled Soils

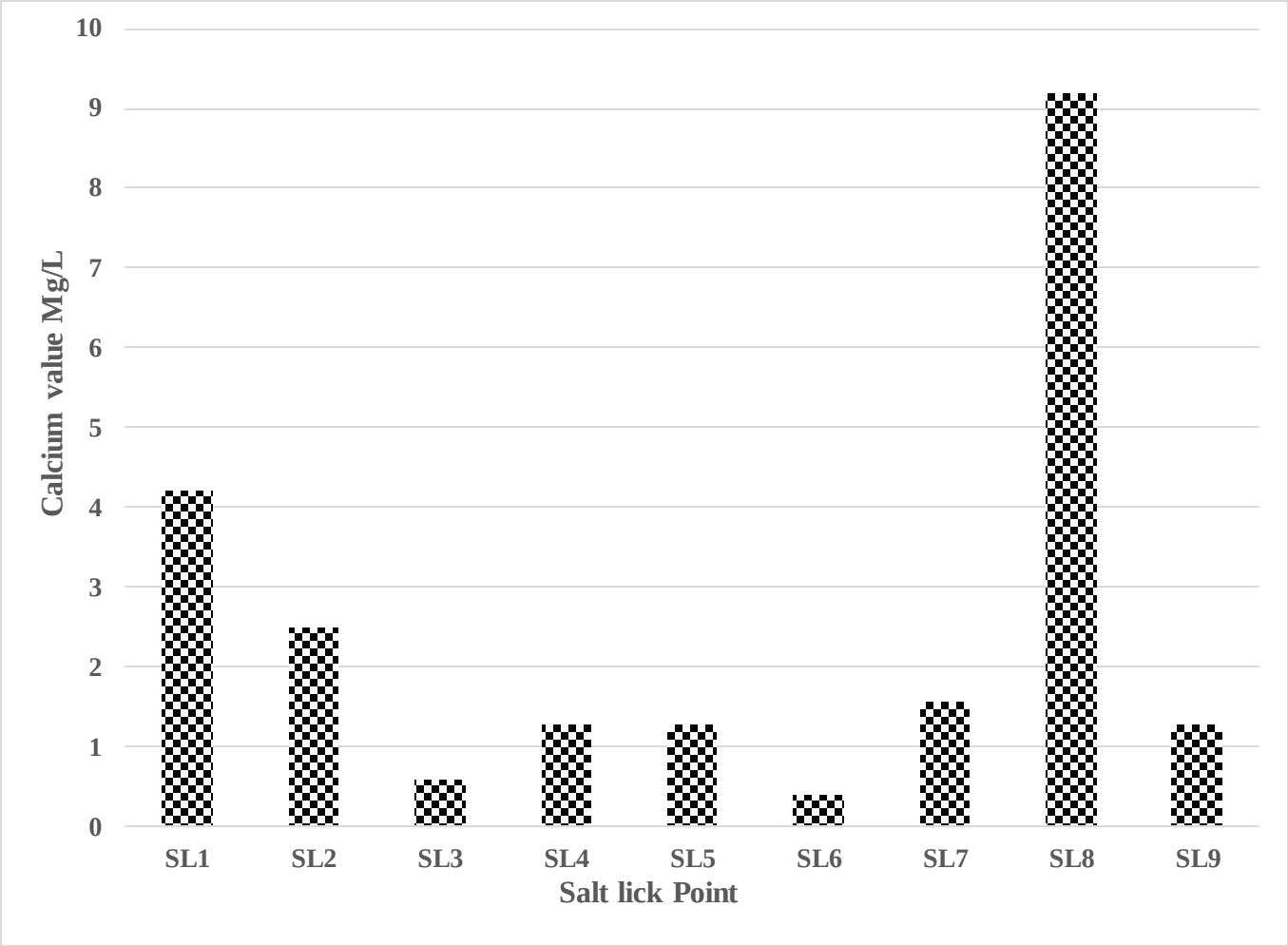


Fig.1: Calcium value (Ca^{2+}) of the soil samples across all saltlick sites

https://dx.doi.org/10.22161/ijeab.51.1 ISSN: 2456-1878

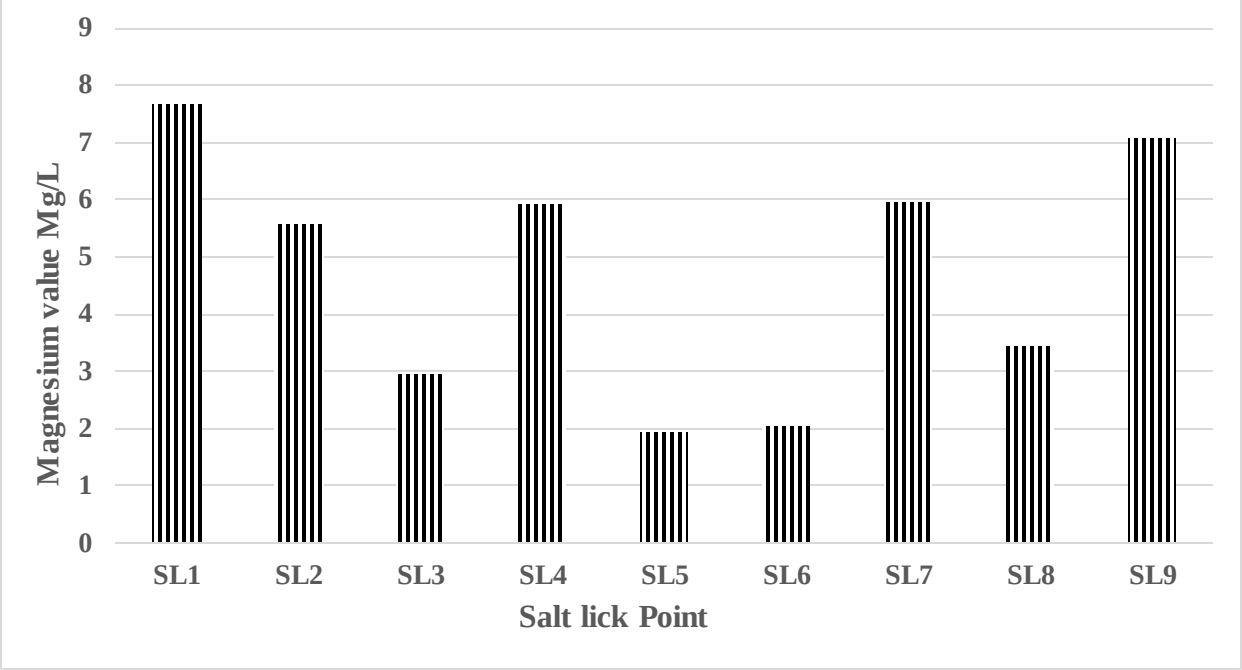


Fig.2: Magnesium value (Mg^{2+}) of the soil samples across all saltlick sites.

Concentration of Trace Elements in the Sampled Soils

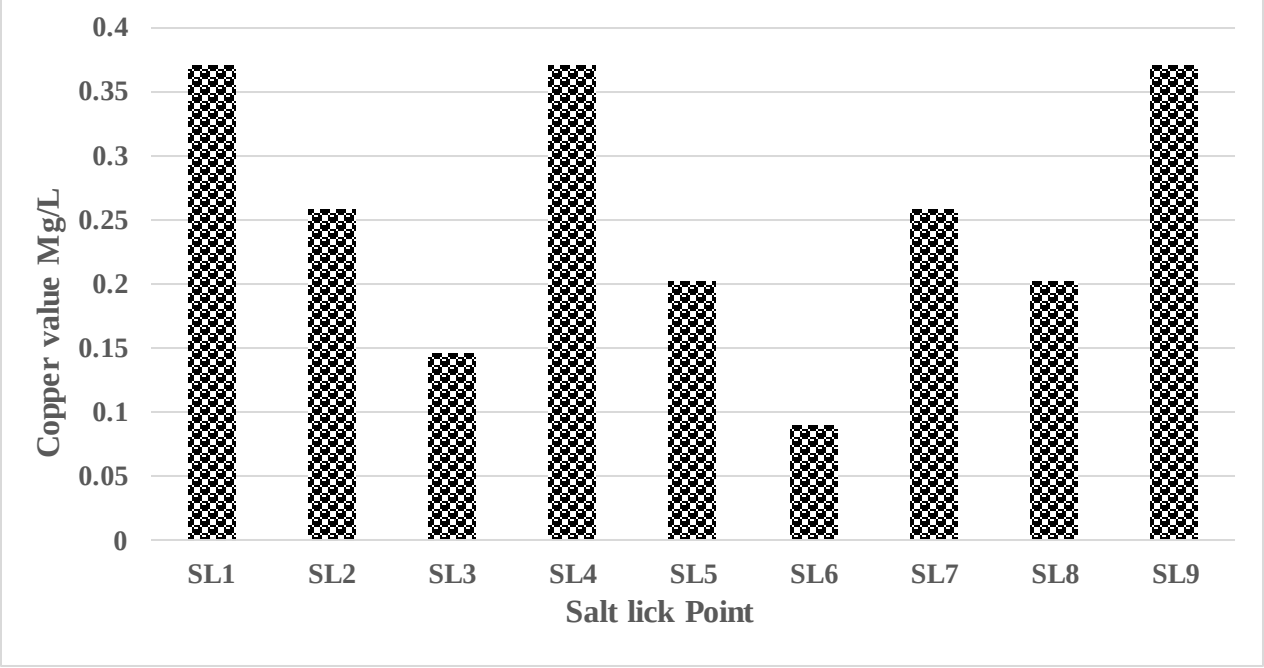


Fig.3: Copper value (Cu^{3+}) of the soil samples across all saltlick sites.

https://dx.doi.org/10.22161/ijeab.51.1 ISSN: 2456-1878

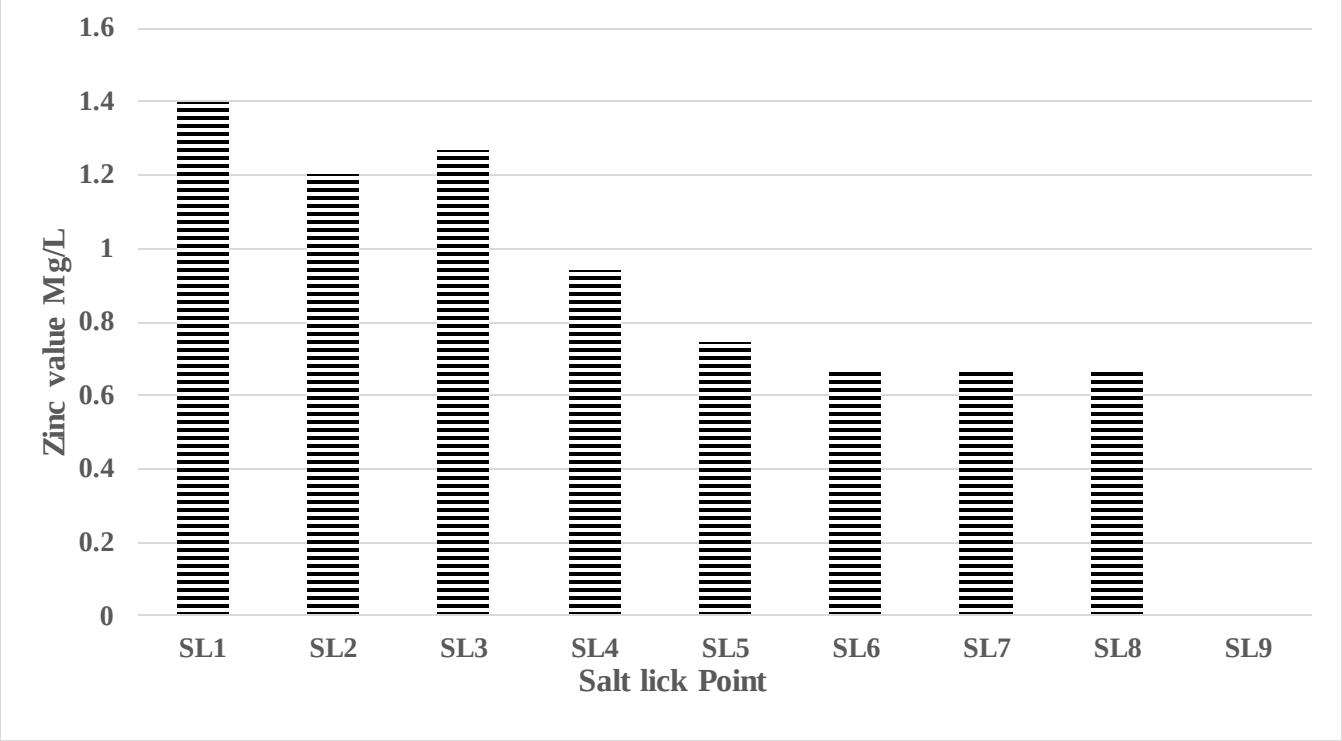


Fig.4: Zinc value (Zn^{2+}) of the soil samples across all saltlick sites.

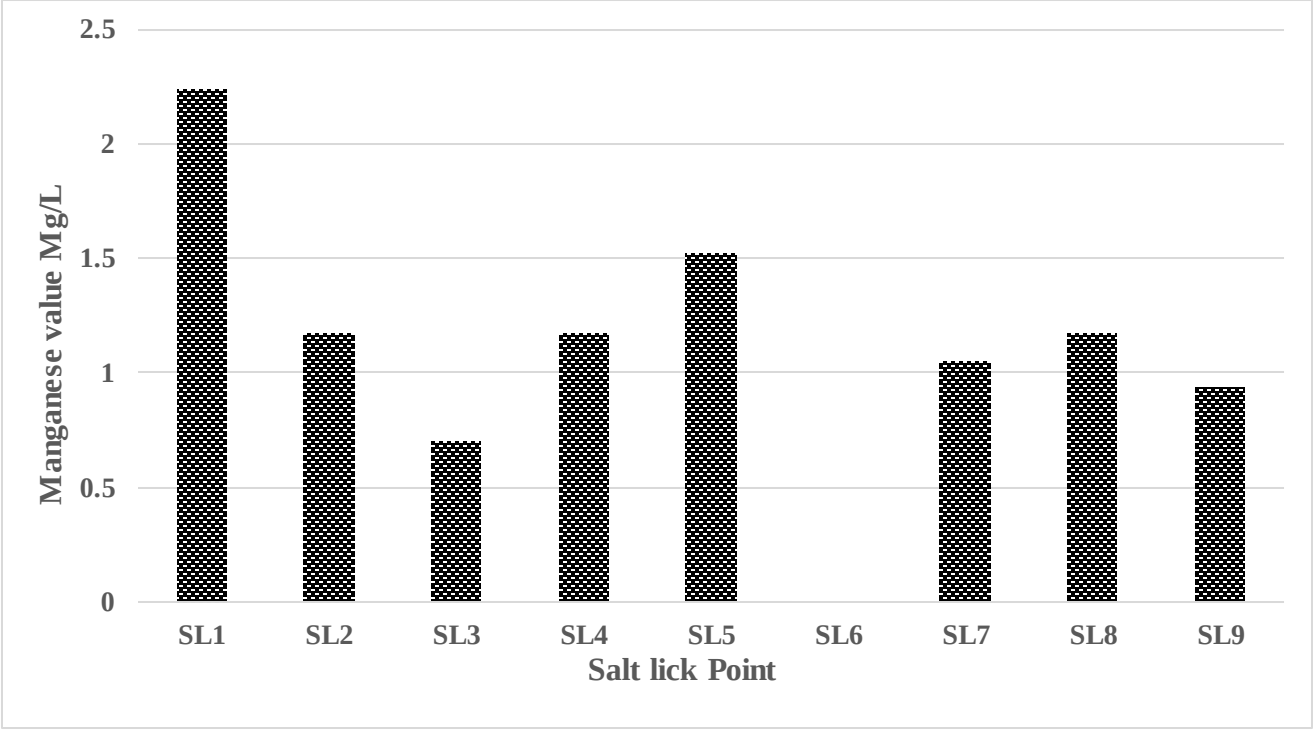


Fig.5: Manganese value (Mg^{2+}) of the soil samples across all saltlick sites.

https://dx.doi.org/10.22161/ijeab.51.1 *ISSN: 2456-1878*

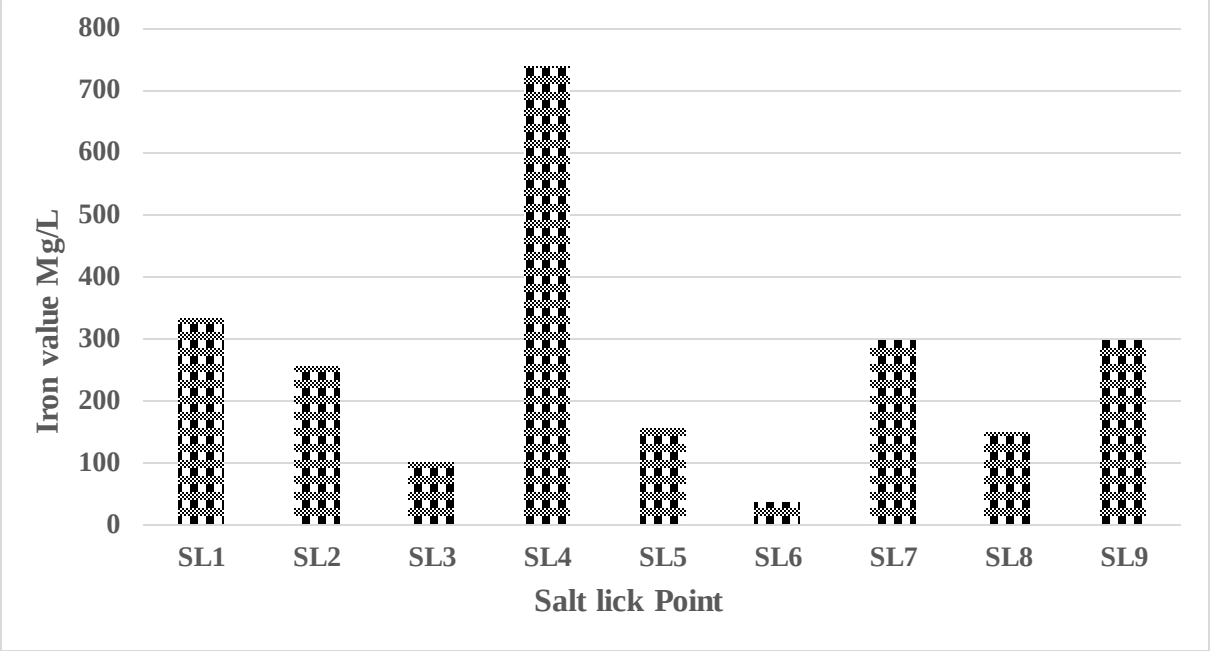


Fig.6: Iron value (Fe^{2+}) of the soil samples across all saltlick sites.

Table 2: Level of Utilization of sampled salt lick sites.

Saltlick Point	Wet season	Dry season
SL1	Xxx	Xx
SL2	Xxx	Xxx
SL3	Xxx	X
SL4	Xxx	X
SL5	Xxx	Xx
SL6	Xxx	Xx
SL7	Xxxx	Xxx
SL8	Xxxx	Xxx
SL9	Xxx	Xxx

KEY: XXXX = Very high, XXX = High, XX= Moderate X= Low, SL= Salt Lick.

Table 3: Tourists' influx in Kainji Lake National Park for the year 2007-2017.

S/N	Year	Nigerians (Domestic)	Foreigners (International)	Total
1	2007	4794	43	4837
2	2008	4025	67	4092
3	2009	4852	25	4811
4	2010	6054	49	6103
5	2011	4677	16	4693
6	2012	3422	07	3429
7	2013	7725	05	7730
8	2014	929	-	929
9	2015	3066	10	3076
10	2016	2993	05	2998
11	2017	192	-	192

IV. DISCUSSION

The important role mineral elements play in the nutrition of wild games cannot be overemphasized as much as the trace minerals required for supplementing their diet. Enzymes are activated by trace elements known as metallo-enzymes. The trace elements are required in minute quantity in animal's diet. The ingestion and assimilation of minerals that are not balanced, nutrient deficient or excessively high in a particular mineral element induce changes in the activities, function or concentration of such element in the body tissue/fluid.

The summary of the mineral content in all the sampled salt lick sites are presented in Table 1. It was observed that the amount of copper found in salt lick 1, 4 and 9 is relatively high 0.37, 0.37 and 0.37 respectivelywhile salt lick 6 has the lowest copper content 0.090 as presented in Figure 3. Observation revealed that the amount of magnesium is high in salt lick 1 (7.69), while the lowest amount is found in salt lick 5 (1.97), as presented in figure 2. Manganese was also found to be high in salt lick 1 (2.24), while the lowest was recorded in salt lick 3 (0.70)however the content in salt lick 6 was not detected as presented in figure 5. Iron concentration is high in salt lick 1 (332.33), while the lowest was recorded in salt lick 6 (36.36) as presented in Figure 6. Zinc has the highest content in salt lick 1 (1.41), and the leastamount was recorded in salt lick 6, 7 and 8 respectively while no mineral content was detectedin salt lick 9, as presented in Figure 4. The content of calcium is highest in salt lick 1 (4.22), with the least recorded in salt lick 6 (0.39) as shown in Figure 1.Observation revealed that the level of utilization of salt licks corresponds to the availability of mineral deposits during both seasons of the year. The calcium concentration was high in salt lick 8, which yielded a high level of utilization while salt lick 3 and 4 were least utilized during the dry seasons. This could summarize the fact that Copper, Iron and Zinc (Trace elements) are not readily released for consumption without an external force of rain. The mineral content of salt lick sites can be a limiting factor to its utilization by fauna resources which has overall effect on wildlife viewing at such sites.During the study, it was observed that, turn-out of tourists visiting the park fluctuates year by year as presented in Table 3. This could be associated to the insecurity status of the country, economic melt-down and the operatingseasons of the park. However, tourists' visitation to salt lick sites and ecological sites is impaired due to a number of reasons as identified during the study. The anthropogenic activities of park intruders is experiencing a rapid increase which can be attributed to decline in fauna population within the sites and in the park at large. This has affected the ecotourism activities of the park to eco-destinations that attract large wildlife populace. Above all, the benefitsderived from salt licks for wildlife health, majorly herbivores are crucial in maintaining a healthy wildlife community for reproduction and survival.

V. CONCLUSION

Salt licks are key places and locations for the ecological dynamics of wildlife communities in protected areas. Salt lickis a natural mineral deposit area where animals visit frequently and actively for mineral uptake through licking.Ecotourism is a special type of wildlife viewing which centers more on a diverse range of fauna species utilizing this site of great potential.The findings showed that facilities and promotion on salt licks are lacking in Nigeriawhich indicates poor conservation of this natural landscape. Salt licks in remote natural areas need to be sustained to protect them against human activities of vegetation loss and species decline. It is evident that interest to visit salt licks is behind the satisfactory level due to the underutilized ecotourism resources of the park. Management of these resources needs sound and effective monitoring as well as evaluation of the fauna resources to sustain the ecological sites. Awareness on the importance of salt licks for ecotourism development need to be enhanced by the park management so as to combat the negative use of these sites for better sustainable conservation measures.

REFERENCES

[1] Abrahams PW (1999)The chemistry and mineralogy of three savannas lick soils.*J. Chem. Ecol., 25:*2215-2228.
[2] Ashley C (2003)Theindian ocean tsunami and tourism.Overseas development institute. www.odi.org.uk/publications/opinion.
[3] Blake JG, Guerra J, Mosquera D, Torres R, Loiselle BA, RomoD (2010) Use of mineral licks by white-bellied spider monkeys (*Atelesbelzebuth*) and red howler monkeys (*Alouattaseniculus*) in eastern Ecuador. *Int. J. Primatol.31*: 471-483.
[4] Dormaar, JF, Walker BD (1996) Elemental content of animal licks along the eastern slopes of the Rocky Mountains in southern Alberta, Canada. Canadian Journal of Soil Science 76: 509–512.
[5] Heimer WE(1974)The importance of mineral licks to Dall sheep in interior Alaska and its significance to sheep management: Proceedings of the Third Biennial Wild Sheep and Goat Council Conference 3: 49–63.
[6] Izawa K(1993)Soil-eating by *Alouatta and Ateles*. *Int. J. Primatol.*, 14: 229-242.

[7] Rea RV,Hodder DP, Child KN(2004) Considerations for natural mineral licks used by moose in land use planning and development. Alces 40: 161–167.
[8] Simmons NM(1982) Seasonal ranges of Dall's sheep, Mackenzie Mountains, Northwest Territories. Arctic 35: 512−518.
[9] Stankowich T (2008) Ungulate flight responses to human disturbance: a review and meta-analysis. Biological Conservation 141: 2159–2173.
[10] Tobler MW,Carrillo-Percastegui SE, Powell G (2009) Habitat use, activity patterns and use of mineral licks by five species of ungulate in south-eastern Peru.*J. Trop. Ecol.* 25:261-270.
[11] Watts TJ, SchemnitzSD(1985) Mineral lick use and movement in a remnant desert bighorn sheep population. Jour. of Wildlife Mgt. 49: 994–996.
[12] Ijeomah HM, Alarape AA,Ogogo AU(2007) Management ethics and strategies towards sustainable tourism development in Jos Wildlife Park. Jour.of Environ.Extens.6:100-106http://www.ajol.info.
[13] NTDC (2005) Nigeria: Beauty in diversity. Nigerian Tourism Development Corporation. Abuja.
[14] Newsome D, Dowling R, Moore S (2005) Wildlife tourism channel view publication clevendon U.K. of foreign investment. "Hum. Ecol., 33(2): 217-244.
[15] Stephenson J D, Mills A, Eksteen, J. J., Milewski, AV, Myburgh JG (2011) Geochemistry of mineral licks at Loskop Dam Nature Reserve, Mpumalanga, South Africa. Environmental Geochemistry and Health, 33: 49–53.
[16] Yinus E (2003)Sustainable tourism and poverty alleviation, World Tourism Organization, Spain, Pp.1-4. **http://www.world-tourism.org.**

https://dx.doi.org/10.22161/ijeab.51.2 *ISSN: 2456-1878*

Forecasting of River Sediment Amount using Machine Model

Fatih ÜNEŞ[1], A. Burhan KARAEMİNOĞULLARI[1], Bestami TAŞAR[1]

[1] Department of Civil Engineering, Iskenderun Technical University, Turkey
Corresponding author. Email: bestami.tasar@iste.edu.tr

Abstract— *Accurate estimation of sediments is important in river structures. The amount of suspended sediments is mostly determined by measurements from observation stations, sediment key curve, artificial intelligence modeling methods. In this study, the estimation of the sediments content was performed by using hydro-meteorological parameters such as river flow, air temperature and precipitation measured between 2011 - 2017 at Omaha Station in Nebraska. For the estimation of sediments amount, Support Vector Machines (SVM) and Generalized Regression Neural Network (GRNN) methods were used. These models were compared by using correlation coefficient (R), mean absolute error (MAE) and root of mean square errors (RMSE). When the measurement and model results were compared, SVM and GRNN models gave consistent results in the estimation of sediments content in rivers. Nevertheless, the SVM method showed slightly better correlation and lower error performance than the GRNN method.*

Keywords— ***Sediment, Prediction, Support vector machine, Generalized regression neural network.***

I. INTRODUCTION

In water resources engineering; Accurate estimation of sediment transported in rivers is of particular importance for the design and planning of river structures. Sediments such as rock fragments, gravel and sand carried by rivers are formed by scraping from the river basin or river bed. The sediment movement is complex and differs according to the topography, geological condition and flow characteristics of the basin. Determining the amount of sediment transported in the regulation of transportation network operations such as flood control and transportation in determining the reservoir volume, selection of water intake and type is an important engineering study. If not taken into consideration; It reduces the capacity of the hopper, leads to clogging of the mouth of the intake structure and shortens the economic life of the plants and leads to material losses. Therefore, accurate sediment observations are directly proportional to the development of soil and water resources.

It is not easy to determine because the amount of sediment varies according to many parameters. It is observed that non-linear functions are formed in the complex structure and appropriate and economical methods are used to solve them.

Usually, the amount of sediment is determined by measurements from field observation stations. Although the measured values from the station give healthy results, they are important in terms of time and cost. even, In some rivers, when the flow rate decreases, sediment measurement is not possible. For these reasons, the estimation of sediment amount is needed in the design of water structures.

In the last years, the artificial intelligence approaches are a technique widely used in water resources engineering and hydrology [1-17].

Thangaraj and Kalaivani [18] estimated the water level in the river using support vector machines. Lafdani et al. [19] investigated their capabilities using Artificial Neural Network (ANN) models to estimate the amount of suspended sediment (SSL) per day in their articles. Afan et al. [20], artificial neural networks (ANN) have been used in their studies to estimate the amount of sediment daily. For this purpose, two different ANN algorithms, feed-forward artificial neural network (FFNN) and radial based function (RBNN) used. They used daily sediment and flow data from the Rantau Panjang station on the Johor River. The results predicted the amount of sediment by the data generated by producing daily flow and sediment time series. When comparing the results, they showed that the FFNN model outperformed the RBNN model in predicting daily sediment.

Taşar et al. [21], in the their articles, artificial neural networks (ANN), M5 tree (M5T) approaches and Multiple Linear Regression (MLR), Sediment rating curve (SRC) using statistical approaches such as daily temperature and flow rate using the estimated daily suspended sediment. The daily data they used was obtained from Iowa station in the USA. They compared these estimation methods with

https://dx.doi.org/10.22161/ijeab.51.2 *ISSN: 2456-1878*

mean square errors (MSE), mean absolute error (MAE) and correlation coefficient (R) according to three statistical criteria. When comparing the results, they observed that the ANN approach had better sediment prediction than other methods.

Yilmaz et al. [22], have tested various regression models to calculate the amount of sediment in two stations (in Turkey-Coruh). Choubin et al. [23], tried to estimate the amount of sediment using hydro-meteorological data.

They used artificial intelligence method models such as Artificial Neural Networks (ANN) and Adaptive Neural Fuzzy Inference System (ANFIS) for sediment time series modeling.

II. STUDY AREA

In this study, support vector machine (SVM) Generalized Regression Neural Network (GRNN) were used for prediction of sediment on Missouri river. The study area was selected as the Douglas county region in the State of Nebraska in the United States. In this study, data were obtained from Omaha station for near 6 years of water year measurements and data were obtained from USGS [24] and US climate data [25]. Figure 1 shows the general view of the Missouri River at Omaha station and the views of the selected area.

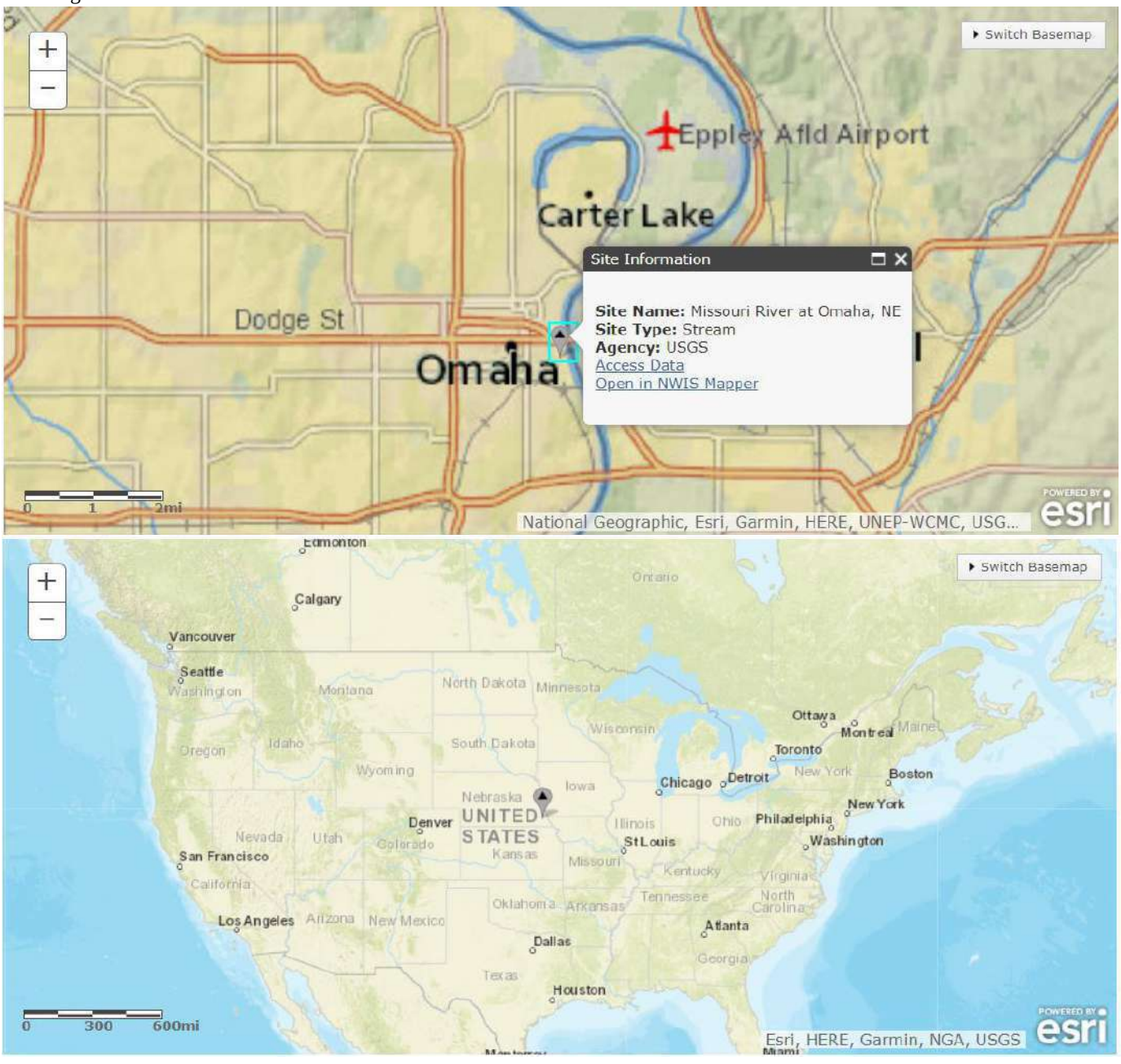


Fig.1: Missouri River at Omaha station [24]

https://dx.doi.org/10.22161/ijeab.51.2 *ISSN: 2456-1878*

III. METHODS

In this study, estimation of sediment amount was made by using hydro-meteorological parameters such as river flow, air temperature and precipitation measured between 2011-2017 at Omaha Station in Nebraska state of Missouri river. Support Vector Machines (SVM) and Generalized Regression Neural Network (GRNN), which are among the artificial intelligence methods, were used to estimate the sediment amount.

3.1 GENERALIZED REGRESSION NEURAL NETWORK (GRNN)

The GRNN method is a supervised artificial neural network model that is radial-based and usually works as an estimator. The strengths of this algorithm are that they produce consistent and fast results and are easy to model. In this artificial neural network model, one neuron is held in the pattern layer for each sample data in the training data set. Therefore, in studies where the training data set is too high, the layer structure grows in direct proportion to the number of sample data, increasing the number of processes and memory requirement.

3.2 SUPPORT VECTOR MACHINES (SVM)

Founded by Vladimir Vapnik and Alexey Chervonenkis [26] in 1963, Support Vector Machines (SVM) is a supervised learning algorithm based on statistical learning theory. It is mainly used to separate data from two classes in an optimal way. For this purpose, decision limits or in other words hyperplanes are determined. Today, SVMs are used in many classification problems ranging from face recognition systems to sound analysis, from water resource engineering to stock market calculations. In this study, SVM method with Poly Kernel function is used.

IV. METHOD RESULTS

In this section, sediment estimation results of Support Vector Machines (SVM) and Generalized Regression Artificial Neural Network (GRNN) methods are examined. Correlation coefficient (R), root mean square error (RMSE) and mean absolute error (MAE) were calculated in the evaluation of the results of the models.

The correlation coefficient (R) measures the strength of the linear correlation between the binary values x and y. R value closest to 1 is the most logical and appropriate. Mean absolute error (MAE) measures the average error magnitude. RMSE and MAE are used to diagnose the probability of errors. RMSE, MAE can go from zero to infinity. Lower values mean more useful.

Expressions of the statistical criteria used in the study are given in equations 1-3.

$$R = \sqrt{\left(\frac{n\Sigma xy - (\Sigma x)(\Sigma y)}{\sqrt{(n(\Sigma x^2) - (\Sigma x)^2)}\sqrt{(n(\Sigma y^2) - (\Sigma y)^2)}}\right)} \quad (1)$$

$$\mathrm{MAE} = \frac{1}{n}\sum_{j=1}^{n}\left|S_{\mathrm{MEASURE}} - S_{\mathrm{ESTIMATION}}\right| \quad (2)$$

$$\mathrm{RMSE} = \frac{1}{n}\sqrt{\sum_{i=1}^{n}\left(S_{\mathrm{MEASURE}} - S_{\mathrm{ESTIMATION}}\right)^2} \quad (3)$$

Here, n is the number of data and S is the daily suspended sediment / sediment amount, concentration (mg / L).

4.1 GRNN METHOD RESULTS

In GRNN analyze, input and output data files were created by using daily measurement data flow, precipitation, temperature and time series. GRNN model was created by applying testing and training phases. The sediment amount, daily precipitation, flow rate and temperature, which consist of 2139 daily observations used in training phase. The models created in the second step were applied to the inputs of the test data generated from the 535 day observations and the results obtained with the model were compared with the measured values. For GRNN method, scatter and distribution graphs of the test graphs of this method are shown in Figure 2, Figure 3, respectively.

https://dx.doi.org/10.22161/ijeab.51.2 ISSN: 2456-1878

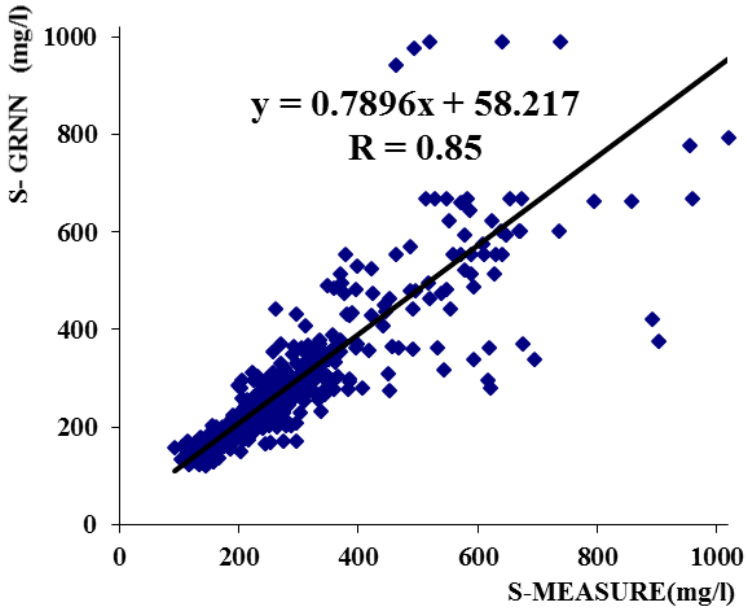


Fig. 2: Measure and GRNN scatter graph for suspended sediment amount test data

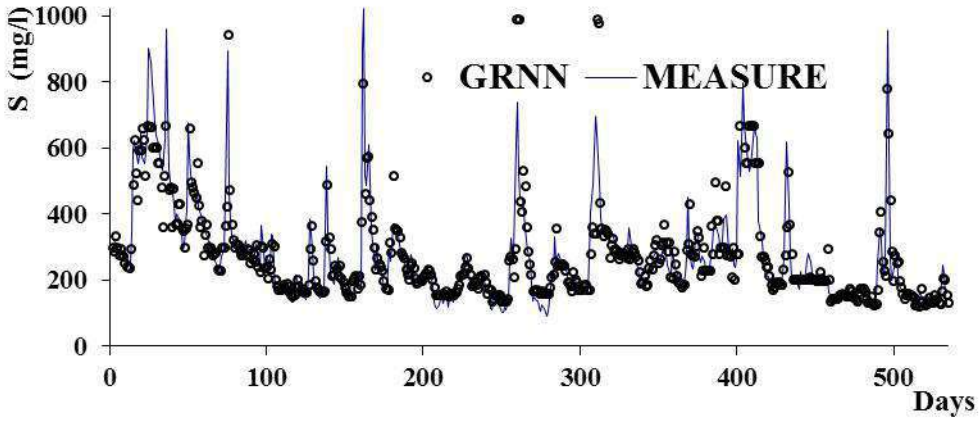


Fig. 3: Measure and GRNN distribution graph for suspended sediment amount test data

Figure 3. shows that distribution of GRNN model test results are quite close to observed values of sediment for the study area. As it is seen in Figure 2, correlation coefficient is calculated as 0.85 for test set of GRNN method.

4.2 SVM METHOD RESULTS

In SVM analyze, input and output data files were created by using daily measurement data flow, precipitation, temperature and time series in SVM model application. And suspended sediment estimation was made using SVM model. The data sets used in the GRNN model were also used in the SVM model. 2139-day observation data were used in the training phase and 535-day observation data were applied to the model. The model results were compared with the field measured values and the model was evaluated. For SVM method, scatter and distribution graphs of the test graphs of this method are shown in Figure 4, Figure 5, respectively

https://dx.doi.org/10.22161/ijeab.51.2 ISSN: 2456-1878

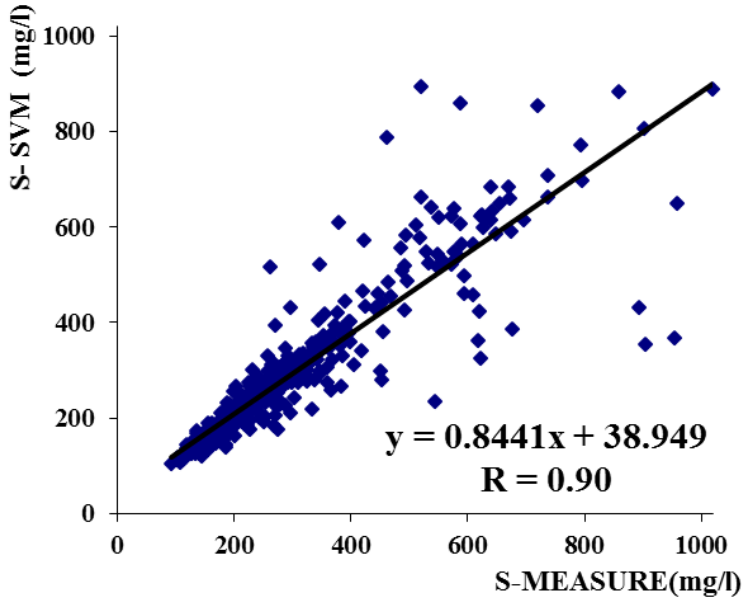


Fig. 4: Measure and SVM scatter graph for suspended sediment amount test data

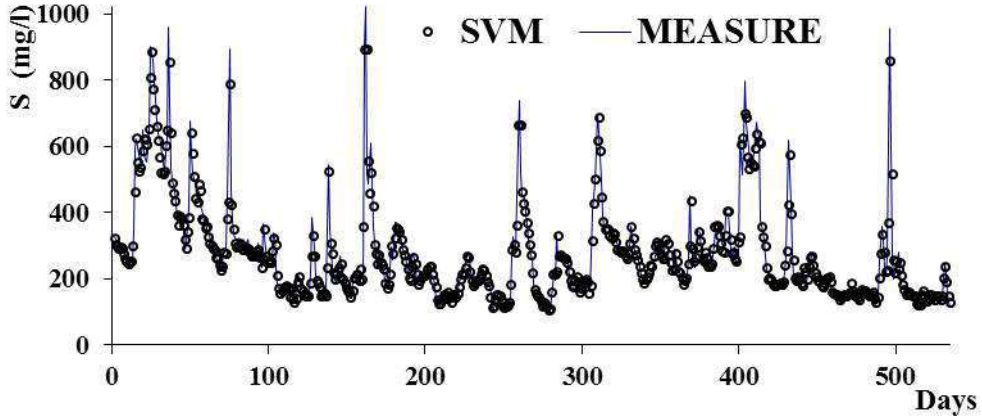


Fig. 5: Measure and SVM distribution graph for suspended sediment amount test data

Results of SVM model show that the correlation coefficient is high and the sediment estimations are closer to the actual values shown in Figure 5. Correlation coefficient is calculated as 0.90 for SVM results as it is seen in Figure 4. The result parameters of the MAE, RMSE and R obtained from the test data will be shown in table form. The results will be used to compare estimations and performance. The statistical results of the models are given in Table 1

Table 1: MAE, RMSE and R parameter results obtained for test data of GRNN and SVM model

Methods	**Method inputs**	**MAE** (Mg/L)	**RMSE** (Mg/L)	**R**
GRNN	Q_t, Q_{t-1}, P_t, P_{t-1}, T_t, S_{t-1}	39.80	81.53	0.85
SVM	**Q_t, Q_{t-1}, P_t, P_{t-1}, T_t, S_{t-1}**	**24.38**	**55.61**	**0.90**

MAE: Mean absolute error, RMSE: Root mean square error, R: Correlation coefficient.
Q (t): Daily flow, Q(t-1): Daily flow time series, P (t): Daily precipitation,
P (t-1): Daily precipitation time series, T (t): Daily temperature, S (t - 1): Daily sediment time series.

According to Table 1, when MAE, RMSE and R statistical criteria were compared, all models were good. All models are evaluated separately; GRNN (39.80 - 81.53 - 0.85) and SVM (24.38 – 55.61 – 0.90) models were found to perform well. Nevertheless, it is observed that the SVM model has a low error rate with high correlation. In addition, the GRNN model are close to SVM prediction performance. When the results were examined, GRNN and SVM models were found to perform better in sediment estimations.

Nevertheless, the SVM method showed slightly better correlation and lower error performance than the GRNN method.

V. CONCLUSION

In this study, daily suspended sediment amount in Missouri River were estimated by using Support Vector Machine (SVM) and Generalized Artificial Neural Network (GRNN) methods. Since the amount of sediment in the river structures actually contains a large number of parameters, the use of the artificial intelligence models, in the solution of this complex problem has enabled us to obtain the most realistic results.

When the SVM model results were compared to the GRNN model, it was observed that the SVM and GRNN model results were close and good.

Since Support Vector Machine applications analyzed for river structures have a low error level and the observed values are close to the estimates, it can be used as an alternative method for the prediction of sediment concentration compared to the classical methods.

ACKNOWLEDGEMENTS

In this paper, hydrological and meteorological data were obtained from USGS (US Geological Survey). The authors would like to thank USGS staff for the transmission of hydrological Measure data.

REFERENCES

[1] Taşar B., Üneş F., Demirci M., Kaya YZ. Yapay sinir ağları yöntemi kullanılarak buharlaşma miktarı tahmini, *DÜMF Mühendislik Dergisi* 2018; 9(1): 543-551.

[2] Üneş F., Doğan S., Taşar B., Kaya Y., Demirci M. The Evaluation and Comparison of Daily Reference Evapotranspiration with ANN and Empirical Methods. *Natural and Engineering Sciences* 2018; 3(3): Supplement, 54-64.

[3] Kaya, YZ., Taşar, B. Evapotranspiration Calculation for South Carolina, USA and Creation Different ANFIS Models for ET Estimation. *"Air and water – components of the environment" conference proceedings* p. 217-224, Cluj-Napoca Romania, 2019, DOI: 10.24193/AWC2019_22.

[4] Taşar, B., Unes, F., Varcin, H. (2019) Prediction of the Rainfall – Runoff Relationship Using NeuroFuzzy and Support Vector Machines. *2019 "Air and Water – Components of the Environment" Conference Proceedings,* Cluj-Napoca, Romania, p. 237-246, DOI: 10.24193/AWC2019_24.

[5] Tasar B., Kaya YZ., Varcin H., Üneş F., Demirci M. Forecasting of Suspended Sediment in Rivers Using Artificial Neural Networks Approach, *International Journal of Advanced Engineering Research and Science (IJAERS)* 2017; 4(12): 79-84.

[6] Üneş F., Demirci M., Taşar B., Kaya YZ., Varçin, H., Modeling of dam reservoir volume using generalized regression neural network, support vector machines and M5 decision tree models. *Applied Ecology and Environmental Research* 2019; 17(3): 7043-7055.

[7] Üneş F., Bölük O., Kaya YZ., Taşar B., Varçin H. Estimation of Rainfall-Runoff Relationship Using Artificial Neural Network Models for Muskegon Basin, *International Journal of Advanced Engineering Research and Science (IJAERS)* 2018; 5(12): 198-205. http://dx.doi.org/10.22161/ijaers.5.12.28

[8] Kaya, YZ., Üneş, F., Demirci, M., Tasar, B., Varcin, H. Groundwater Level Prediction Using Artificial Neural Network and M5 Tree Models. *Air and water components of the environment conference, 2018*; DOI: 10.24193/AWC2018_23

[9] Demirci M., Taşar B., Kaya YZ, Varçin, H. Estimation of Groundwater Level Fluctuations Using Neuro-Fuzzy and Support Vector Regression Models, *International Journal of Advanced Engineering Research and Science 2018;* 5(12): 206-212. http://dx.doi.org/10.22161/ijaers.5.12.29

[10] Demirci, M., Unes, F., Kaya, YZ., Mamak, M., Tasar, B., Ispir, E. Estimation of groundwater level using artificial neural networks: a case study of Hatay-Turkey. *In 10th International Conference „Environmental Engineering* " 2017, March.

[11] Demirci M., Üneş F., Körlü S. Modeling of groundwater level using artificial intelligence techniques: a case study of Reyhanlı region in Turkey. *Applied Ecology and Env. Research* 2019; 17(2): 2651-2663. http://dx.doi.org/10.15666/aeer/1702_26512663

[12] Kaya, YZ., Üneş, F., Demirci, M., Taşar, B., Varçin, H. Groundwater Level Prediction Using Artificial Neural Network and M5 Tree Models. *Aerul si Apa. Componente ale Mediului, 2018*,195-201.

Üneş F., Demirci M., Mertcan Z, Taşar B., Varçin H., Ziya Y. Determination of Groundwater Level Fluctuations by Artificial Neural Networks. *Natural and Engineering Sciences 2018*; 3(3): Supplement, 35-42.

[13] Üneş F, Maruf AG., Taşar B. Ground Water Level Estimation for Dörtyol region in HATAY. *International Journal of Environment, Agriculture and Biotechnology 2019*; 4(3): 859-864.

[14] Üneş F., Demirci M., Taşar B., Kaya YZ., Varçin H. Estimating Dam Reservoir Level Fluctuations Using Data-

Driven Techniques, *Pol. J. Environ. Stud*. 2019; 28(5): 1-12. DOI: 10.15244/pjoes/93923

[15] Unes F., Gumuscan F. G., Demirci M. (2017). *Prediction of Dam Reservoir Volume Fluctuations Using Adaptive Neuro Fuzzy Approach*. European Journal of Engineering and Natural Sciences , 2 (1) , 144-148 .

[16] Demirci, M., Unes, F., & Akoz, M. S. (2016). Determination of nearshore sandbar crest depth using neural network approach. *International Journal of Advanced Engineering Research and Science*, *3*(12).

[17] Kalaivani, R., & Thangaraj, P. (2012). River water level prediction in satellite images using support vector machine. *International Journal of Computer Applications*, *43*(23).

[18] Lafdani, E. K., Nia, A. M., & Ahmadi, A. (2013). Daily suspended sediment load prediction using artificial neural networks and support vector machines. *Journal of Hydrology*, 478, 50-62.

[19] Afan, H. A., El-Shafie, A., Yaseen, Z. M., Hameed, M. M., Mohtar, W. H. M. W., & Hussain, A. (2015). ANN based sediment prediction model utilizing different input scenarios. *Water resources management*, 29(4), 1231-1245.

[20] Tasar B., Kaya YZ., Varcin H., Üneş F., Demirci M. Forecasting of Suspended Sediment in Rivers Using Artificial Neural Networks Approach, *International Journal of Advanced Engineering Research and Science (IJAERS)* 2017; 4(12): 79-84.

[21] Yilmaz, B., Aras, E., Nacar, S., & Kankal, M. (2018). Estimating suspended sediment load with multivariate adaptive regression spline, teaching-learning based optimization, and artificial bee colony models. *Science of the Total Environment*, *639*, 826-840.

[22] Choubin, B., Darabi, H., Rahmati, O., Sajedi-Hosseini, F., & Kløve, B. (2018). River suspended sediment modelling using the CART model: a comparative study of machine learning techniques. *Science of the Total Environment*, *615*, 272-281.J. Clerk Maxwell, A Treatise on Electricity and Magnetism, 3rd ed., vol. 2. Oxford: Clarendon, 1892, pp.68–73.

[23] USGS.gov | Science for a changing world [WWW Document], n.d. URL https://www.usgs.gov/

[24] U.S. Climate Data. About the climate in the United States. www.usclimatedata.com

[25] Vapnik, V. N., & CHERVONE. AY. (1965). On a class of pattern-recognition learning algorithms. *Automation and Remote Control*, *25*(6), 838.

https://dx.doi.org/10.22161/ijeab.51.3 *ISSN: 2456-1878*

How Reliable are Farmers' Perceptions about Climate Change? A Case Study in the Upper East Region of Ghana

Abdul-Razak Zakaria[1], Kenichi Matsui[2]

[1]Graduate School of Life and Environmental Sciences, The University of Tsukuba, Japan
Email: nurazak830@gmail.com
[2]Faculty of Life and Environmental Sciences, The University of Tsukuba
Email: kenichim@envr.tsukuba.ac.jp

Abstract**—** *The 2014 IPCC report reiterated the importance of local farmers' perceptions about climate change. A growing number of scientists supports that farmers' in-depth understanding of climate change hazards and their active participation in mitigation actions are key to improving adaptation. This paper attempts to analyze smallholder rice farmers' perceptions and knowledge about climate change hazards in the Upper East Region of Ghana mainly by looking at the national climatic data and the results of the questionnaire survey we conducted. The climatic data were further analyzed through the Mann-Kendall trend test to find relations between actual rainfall and temperature changes with farmers' observations. Our analysis on perceptions shows that more than 60% of the respondents experienced climate hazards in the forms of increasing temperature, decreasing rainfall and changing planting time. This result is also supported by the Mann-Kendall trend test. The change in planting time is attributable to the increasing coefficient of variation of the annual rainfall from 16.5% (1996-2005) to 28.1% (2006-2015). It is also due to substantial rainfall deviations within the Region in May, from 1,000 mm in the decade between 1996 and 2005 to 500 mm in the following decade (2006-2015). We argue that farmers' observations are largely reliable particularly in observing changes in rainfall patterns. Their observations can also supplement insufficient local meteorological records to better understand local climate change conditions in Western Africa.*

Keywords**— Climate change, farmer's perceptions, food security, rainfall and temperature patterns.**

I. INTRODUCTION

The African continent is known to be particularly vulnerable to climate change (Boko et al., 2007). Most parts of Africa have experienced rising near surface temperatures by 0.5 ^{o}C or more in the past 50 to 100 years. It is projected that further changes in precipitation, temperature and CO_2 level in the atmosphere will cause serious damage to terrestrial ecosystems and agriculture (Niang et al., 2014).

In Ghana, compelling evidence shows temperature increase and rainfall decrease (Antwi-Agyei 2012; Dietz et al., 2004; Issahaku et al., 2016; Nkrumah et al., 2014, Stanturf et al., 2011). It is also projected that this trend will continue (Amuakwa-Mensah, 2014, Dietz et al., 2004). Many researchers have discussed climate change impacts on agriculture in Ghana and other Sub-Saharan African countries (Makate et al., 2017; Mertz et al., 2008; OECD-FAO, 2016) and noted discrepancy and deficiency in observed rainfall data (UNECA, 2014; Niang et al., 2014).

As a result, a number of researchers have examined farmers' perceptions about climate change impacts to better understand vulnerability, coping capacity and adaptation (Allahyari et al., 2016; Deressa et al., 2010; Fosu-Mensah et al., 2012; Ghosh et al., 2015; Salick and Byg, 2007). The 2014 IPCC report demonstrates the increasing recognition and integration of local farmers' perceptions/awareness about climate change among scientists (IPCC, 2014). Growing evidence supports that farmers' in-depth understanding about climate change hazards is key to improving adaptation (Niang et al., 2014).

Here the fundamental and debatable question arise as to the extent to which farmers' perceptions about climate change are reliable to understand local climatic conditions. To deal with this concern, we attempt to validate farmers' perceptions about climate change by comparing with the actual meteorological data in the same study area for two decades (1996 to 2015).

https://dx.doi.org/10.22161/ijeab.51.3 *ISSN: 2456-1878*

II. RESEARCH METHODOLOGY

2.1 Study area

This research was conducted in five districts of the Upper East Region, one of three northern regions of Ghana. The region is located at 10.7082° N, 0.9821° W and covers an area of 8,842 km2. The population density is 103 persons/km2. Agriculture is the dominant economic activity here. It employs 80% of the population. It is also the second poorest region in Ghana. Its climate is characterized by two distinct seasons: the wet season from May to October, and the dry season from November to April. Mean annual rainfall in the region ranges from 950 mm to 1,100 mm (Ghana Statistical Service, 2014).

The Upper East Region is one of the major rice producing regions in Ghana. Most recent statistics shows that it accounted for about 21% of rice output in the nation (MOFA, 2016). Non-irrigated rice farming is practiced in all thirteen districts in the region, while irrigated rice farming is practiced in only three districts near Tono and Vea dams. Average farm size per household is 1.3 hectares with a low average yield of 1.8 t/ha (compared with the national average of 2 t/ha). Hoe, cutlass and in some cases bullock ploughs and sickle are mainly used for cultivation.

2.2 Sampling

A preliminary survey was carried out to understand the feasibility and significance of the survey for the local participants. Considering the significance of rice production, we selected five out of 13 districts: Bawku West, Binduri, Garu-Tempane, Pusiga and Bawku East. In these districts, we applied stratified random sampling to select sample communities. Using simple random sampling, we selected three representative farming communities in each district and interviewed ten farmers in each community (Table 1). In total, 150 farmers fully participated in our survey. The selected districts have similar characteristics in terms of the climate, soil type, farming system, culture, language and cultivated crops (Ghana Statistical Service, 2014). Due to its similar characteristics, it is mainly known as "Bawku zone."

Table.1: Summary of sampled districts and communities

Districts	District population	Sampled communities	Sample size
Bawku West	94,034	Boya-natinga	10
		Gumbo-natinga	10
		Gozesi	10
Binduri	61,576	Azum-sapeliga	10
		Nayoko	10
		Yalugu	10
Garu-Tempane	130,003	Azimbasi	10
		Baring	10
		Yiziidug	10
Pusiga	57,677	Dabia	10
		Suande	10
		Zong-natinga	10
Bawku East	98,538	Gentiga	10
		Kuka yakin	10
		Tampizua	10
Total	441,828	15	150

(Ghana Statistical Service, 2014)

2.3 Data collection and analysis

In the survey, we first attempted to identify socio-demographic characteristics of the respondents partly to understand whether these characteristics influenced their perceptions. We then asked the respondents to describe their observations about long-term (20 years) temperature, rainfall patterns and changes. They were also asked to note if they had made any changes in planting seasons in response to climate change. As most respondents had limited reading and writing skills in English, the questionnaire was interviewer-administered. Also, to overcome language barrier, we translated the questions into the local language. As a result, we had 100% response rate. Survey responses were coded and analyzed in the statistical package for social sciences (SPSS version 20) worksheet. To ascertain climate change patterns in the selected districts, we gathered annual rainfall and temperature data from 1996 to 2015. These data are available at the meteorological department of the Manga Savannah Research Institute (SARI). We used XLSTAT 2017 software to conduct the analysis. We also applied the coefficient of variation (CV) technique to assess the annual and seasonal rainfall variations.

Several studies have employed the Mann-Kendall trend (Mann 1945; Kendall 1975) and the Sen's estimator (Sen 1968) to analyze rainfall and temperature data (Kiros et al., 2016; Gajbhiye et al., 2016; Kabo-Bah et al., 2016; Merabtene et al., 2016; Partal et al., 2011; Rahmat et al., 2006; Yadav et al., 2014). It is a non-parametric trend test for time series data. It enables us to determine whether there is a trend in the time series data or not. Null hypothesis (H_o) is that there is no trend in the data series. The alternate hypothesis (H_a) is that there is a trend in the data series. The null hypothesis (H_o) is rejected if the p-value is lower than the significance level. However, when the p-value is higher than the significance level, then we accept the null hypothesis (H_o). The result of the Mann-Kendall test, which shows the statistical significance of the trend in the data set, is usually complemented with the Sen's slope estimator, which denotes the magnitude of the

trend. The mathematical representation of the Mann-Kendall trend test is as follows:

The nth time series values denoted as: X1, X2, X3…., Xn are replaced in equation (1) below by their relative ranks, R1, R2, R3…., Rn.

$$S = \sum_{i=1}^{n} \left\{ \sum_{j=i+1}^{n} sgn\,(R_i - R_j) \right\} \qquad (1)$$

where,

$$sgn\,(x) = 1 for\ x > 1$$
$$sgn\,(x) = 0\ for\ x = 0$$
$$sgn\,(x) = -1 for\ x < 0$$

Should null hypothesis (H_o) is true, then S is approximately normally distributed with: $\mu = 0$

$$\delta = n\frac{(n-1)(2n+5)}{18} \qquad (2)$$

For data sample (n) larger than 10, the standard test statistic Z is computed as the Mann-Kendall test statistic as follows:

$$Z = \begin{cases} \dfrac{S-1}{\sqrt{var(S)}} & if\ S > 0 \\ 0 & if\ S = 0 \\ \dfrac{S+1}{\sqrt{var(S)}} & if\ S < 0 \end{cases}$$

The presence of a statistically significant trend is evaluated by using the Z value. Positive values of Z indicate an increasing trend, while negative values show decreasing trends. To test for either an increasing or decreasing monotonic (increasing or decreasing) trend at α level of significance, H_o should be rejected if $|z| > z_{1-\alpha/2}$, where $z_{1-\alpha/2}$ is obtained from the standard cumulative distribution tables.

The Sen's estimator is determined using the equation (3):

$$\beta = Median\frac{x_j - x_i}{j - i}\ for\ all\ i \le j \qquad (3)$$

where; β is the robust estimate of the trend magnitude. A positive value of β indicates an "upward trend," and a negative value indicates a "downward trend" (Yadav et al., 2014; Rahmat et al., 2015).

The coefficient of variation (CV) explains the deviation in a data series from its central tendencies. The coefficient of variability is found by expressing the standard deviation of a data set as a percentage of its mean value (Bari et al., 2017). Precipitation is the most important parameter that shapes hydrology, water quality and vegetation. If the CV value is high, it depicts a larger spatial variation in the data set (Gajbhiye et al., 2016).

III. RESULTS AND DISCUSSION

3.1 Socio-demographic characteristics of respondents

Regarding the socio-demographic characteristics of the respondents, we found that 61% belonged to an age bracket of 30-49 years old. Male respondents consisted of 64%. However, this does not mean that males dominate rice farming in our study area. Actually, we observed that women were predominant workers in rice farming. In terms of education levels, 81% had no formal education. This constituted 89% of the total female respondents and 76% of the total male respondents. The literacy rate of the female respondents was below the estimated 2018 national literacy level of 76% (Countrymeters, 2018). Despite this social setback, almost 70% of the respondents had more than 11 years of experience in rice farming (Table 2). Hence their perceptions about long-term climate variability can be overall compatible to the recorded climate data we use.

Table.2: Socio-demographic characteristics of respondents

Social characteristics	Category	Frequency (%)
Age	20-29	10 (7%)
	30-39	33 (22%)
	40-49	59 (39%)
	50-59	28 (19%)
	60 & above	20 (13%)
Gender	Female	54 (36%)
	Male	96 (64%)
Education	Junior high	15 (10%)
	Senior high	4 (3%)
	Tertiary	7 (5%)
	Non-formal	3 (2%)
	No education	121 (80%)
Years of experience in rice farming	1- 10	47 (31%)
	11- 20	59 (39%)
	21- 30	30 (20%)
	31- 40	9 (6%)
	41-50	4 (3%)
	51- 60+	1 (1%)
Total		150

https://dx.doi.org/10.22161/ijeab.51.3 *ISSN: 2456-1878*

3.2 Respondents' perceptions about climate change

To understand farmers' perceptions about climate change, we attempted to identify respondents' perceptions about changes in temperature, rainfall pattern, planting time and drought frequency. If they observed changes, we then attempted to find out what impacts, if any, they experienced. The respondents stated that temperature (62%) and drought frequency had increased (65%), whereas rainfall had decreased (84%). In response, they had changed planting time (82%). In all five districts of the Bawku zone, 98% of the respondents experienced declining rice yields on their farms (Table 3). Overall, nearly all respondents (98%) believed that climate change and weather-related hazards had reduced rice yields.

Table.3: Gender and farmers' perceptions crosstabulation

Gender and farmers' perceptions	Female	Male	Total
Increasing temperature	25	32	57
	46%	33%	38%
	29	64	93
	54%	67%	62%
Decreasing rainfall	9	15	24
	17%	16%	16%
	45	81	126
	83%	84%	84%
Changing planting time	7	20	27
	13%	21%	18%
	47	76	123
	87%	79%	82%
Increasing drought	25	27	52
	46%	28%	35%
	29	69	98
	54%	72%	65%
Reduced crop yield	1	2	3
	2%	2%	2%
	53	94	147
	98%	98%	98%
Total	54	96	150
	100%	100%	100%

During our field observation, we found different gender roles in producing rice in the study area. We attempted to see if gender difference affected local climate change perceptions. We conducted a chi-squared analysis and found a significant gender difference regarding drought events ($\chi 2$ = 5.038, df = 5 and p = 0.025) (Table 4). Whereas 72% of the males perceived increasing drought events, 54% of the females did so (Table 3). Although we cannot identify a definite reason to explain this gender disparity, we observed that the male respondents visited their farms more often than their female counterparts largely because the women had to attend to household chores and childcare.

Table.4: Socio-demographics and farmers' perceptions

	Increasing temp.	Decreasing rainfall	Changing planting time	Increasing drought	Reduced crop yield
Age	0.17	4.69	2.78	4.35	5.77
	(0.99)	(0.32)	(0.59)	(0.36)	(0.22)
Gender	2.46	0.03	1.45	5.04	0.01
	(0.12)	(0.86)	(0.23)	(0.03*)	(0.92)
Education	0.71	2.34	12.34	0.33	0.73
	(0.95)	(0.67)	(0.02*)	(0.98)	(0.95)
Experience	2.916	10.044	5.665	5.212	2.982
	(0.71)	(0.74)	(0.34)	(0.39)	(0.70)

Note: Numbers in parentheses denote the p-value.

3.3 Meteorological data analysis

We examined the extent to which the above findings correspond with the meteorologically recorded rainfall and temperature data in the five districts. The lowest and highest annual rainfall were 671 mm and 1,562 mm between 1996 and 2015. The p-value of 0.048 showed a decreasing trend in annual rainfall in the area (Table 5). The Sen's slope value of -14.572 suggested that rainfall decreased at a rate of about 15% annually in the past twenty years.

Similarly, the highest and lowest temperature were 37.2 ^{o}C and 20.5 ^{o}C between 1996 and 2015. The p-value for the maximum temperature of 0.003 means that there existed a trend in the data set. However, the p-value for the minimum temperature of 0.795 shows no trend in the minimum temperature data set (Table 6). The Sen's slope indicates that the maximum temperature increased by 0.055. It also indicates that the minimum temperature decreased by 0.011. The decrease in minimum temperature, however, was not significant.

Considering the reliability of farmers' perceptions about increasing temperature and drought events, decreasing rainfall and changing planting time, we conducted the Mann-Kendall trend test. The result showed positive relations. This implies that the respondents' perceptions, especially more experienced ones, sufficiently demonstrated a reliable understanding and knowledge about changes in multiple climate conditions in the area. These results positively correspond with previous related studies (Kabo-Bah et al., 2016; Ofori-Sarpong, 2001; Zampaligré et al., 2014; Gbetibouo, 2009; Fosu-Mensah et al., 2012).

https://dx.doi.org/10.22161/ijeab.51.3 *ISSN: 2456-1878*

To understand the validity of farmers' responses to climate change by changing planting time, we calculated and examined two decades coefficient of variation in annual rainfalls in two ten-year periods: 1996-2005 and -2006-2015. The results were 16.5% and 28.1%, respectively. This result means that the average annual rainfall in the area varied substantially between 2006 and 2015 by 28%. Also, a graphical plot of the monthly rainfall data in the study area between 1996-2005 and 2006-2015 revealed a substantial shift in the timing and amount of rainfall. In the first decade, maximum attainable rainfall was 1,000 mm whereas the second decade experienced a maximum rainfall of 500 mm in May (Figures 2 and 3). Generally, however, the growing season in this area starts from May and ends in October (Ghana Statistical Service, 2014).

Table.5: Trend test for average annual rainfall (mm)

Mann-Kendall trend test / Two-tailed test (Average Annual rainfall (mm)	
Kendall's tau	-0.326
S	-62.000
Var(S)	950.000
p-value (Two-tailed)	0.048
Alpha	0.05

Table.6: Trend test for average annual maximum and minimum temperatures (°C)

Mann-Kendall trend test/Two-tailed test (Maximum temperature)		Mann-Kendall trend test/Two-tailed test (Minimum temperature)	
Kendall's tau	0.495	Kendall's tau	0.047
S	94	S	-9
Var (S)	950	Var (S)	949
p-value (Two-tailed)	0.003	p-value (Two-tailed)	0.795
Alpha	0.05	Alpha	0.05

IV. CONCLUSION

This paper has demonstrated that smallholder rice farmers in the Upper East Region of Ghana have carefully observed multiple climate change events and responded to these challenges. More than 60% of the respondents perceived increasing temperature and drought frequency as well as decreasing rainfalls (84%). Nearly all respondents (98%) experienced crop yield reduction due to climate-related hazards. In response, 82% of our respondents changed their planting time.

Their climate observation and remedial actions substantially corresponded with the meteorologically recorded climate data. From 2006 to 2015, the trend of decreasing rainfall in the study area became severe. Rainfall decreased at the rate of about 15% annually. The detected shift in the monthly rainfall decreased from 1,000 mm in the 1996-2005 period to 500 mm in the 2006-2015 period.

Farmers' knowledge about changing climate conditions is based on their careful daily observation. Local farmers, especially experienced ones, can help scientists and policymakers better understand increasingly localized climate events and hazards. Their knowledge can better inform climate-related decision making as well as food security policies in Ghana and Sub-Saharan Africa at large. In turn, policymakers' better understanding about farmers' capacities and needs will make agricultural policies more climate adaptive in the future.

ACKNOWLEDGEMENT

We are very grateful to the Manga Savannah Agricultural Research Institute and 150 farmers who cooperated with us and provided information for this research.

REFERENCES

[1] Allahyari, M. S., Ghavami, S., and Masuleh, Z. D. (2016). Understanding Farmers' Perceptions and Adaptations to Precipitation and Temperature Variability: Climate, 4 (58), 1-16. https://doi.org/10.3390/cli4040058.

[2] Antwi-Agyei, P. (2012). Vulnerability and adaptation of Ghana's food production systems and rural livelihoods to climate variability. Ph.D. diss., University of Leeds, School of Earth and Environment. http://etheses.whiterose.ac.uk/5044/. Viewed on June 25, 2018.

[3] Antwi-Agyei, P., Stringer, L. C., and Dougill, A. J. (2014). Livelihood adaptations to climate variability: Insights from farming households in Ghana. Regional Environmental Change, 14 (4), 1615-1626. https://doi.org/10.1007/s10113-014-0597-9.

[4] Boko, M., Niang I., Nyong A., Vogel C., Githeko A., Medany M., Osman-Elasha B., Tabo R., and Yanda P. (2007). Africa. Climate Change 2007: Impacts, Adaptation and Vulnerability. Contribution of Working Group II to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. Eds: Parry, M. L., Canziani, O.F., Palutikof, J. P., van der Linden, P. J. and Hanson, C. E. Cambridge University Press, Cambridge UK, pp. 433-467.

[5] Connolly-Boutin, L., and Smit, B. (2016). Climate change, food security, and livelihoods in sub-Saharan Africa. Regional Environmental Change, 16 (2), 385-399. https://link.springer.com/article/10.1007/s10113-015-0761-x

[6] Countrymeters. (2018). Ghana Population. Countrymeters.info/en/Ghana. http://countrymeters.info/en/Ghana#literacy. Viewed on March 5, 2018.

[7] Fosu-Mensah, B. Y., Vlek, P. L. G., and Mac Carthy, D. S. (2012). Farmers' perception and adaptation to climate change: a case study of Sekyedumase district in Ghana. Environment, Development and Sustainability, 14 (4), 495-505. https://doi.org/10.1007/s10668-012-9339-7.

[8] Gajbhiye, S., Meshram, C., Mirabbasi, R., and Sharma, S. K. (2016). Trend analysis of rainfall time series for Sindh river basin in India. Theoretical and Applied Climatology, 125 (34), 593-608. https://doi.org/10.1007/s00704-015-1529-4.

[9] Gbetibouo, G. A. (2009). Understanding farmers' perceptions and adaptations to climate change and variability: The case of the Limpopo Basin, South Africa (Vol. 849). Intl Food Policy Res Inst.

[10] Ghana Statistical Service-Bawku Municipality. (2014). District Analytical Report–Bawku Municipality. Population and Housing Census, 1-70. Retrieved from http://www.statsghana.gov.gh/docfiles/2010_District_Report/Upper East/BawkuMunicipality.pdf. Viewed on January 14, 2020.

[11] Ghana Statistical Service-Pusiga. (2014). District Analytical Report-Pusiga District. Population and Housing Census, 1-71. Retrieved from http://www.statsghana.gov.gh/docfiles/2010_District_Report/Upper East/Pusiga.pdf. Viewed on January 14, 2020.

[12] Ghana Statistical Service- Bawku West. (2014). 2010 Population and Housing Census Analytical Report: Bawku West District. Ghana Statistical Service, 1-75. Retrieved from https://new-ndpc-static1.s3.amazonaws.com/CACHES/PUBLICATIONS/2016/06/06/Bawku+West+2010PHC.pdf. Viewed on January 14, 2020.

[13] Ghana Statistical Service-Garu-Tempane. (2014). District Analytical Report-Garu-Tempane District. Population and Housing Census, 1-79. Retrieved from http://www2.statsghana.gov.gh/docfiles/2010_District_Report/Upper%20East/GARU%20TEMPANE.pdf. Viewed on January 14, 2020.

[14] Ghana Statistical Service-Binduri, (2014). District Analytical Report - Binduri District. Population and Housing Census, 1-70. Retrieved from http://www2.statsghana.gov.gh/docfiles/2010_District_Report/Upper%20East/BINDURI.pdf. Viewed on January 14, 2020.

[15] Ghosh, B. C., Osmani, M. A. G., and Hossain, M. E. (2015). Perception of and adaption capacities to climate change adaption strategies by the rice farmers: A case of Rajshahi District in Bangladesh. Journal of Economics and Sustainable Development. 6 (2),136-145.

[16] Issahaku, A., Campion, B. B., and Edziyie, R. (2016). Rainfall and temperature changes and variability in the Upper East Region of Ghana. Earth and Space Science. 3, 284-294. https://doi.org/10.1002/2016EA000161

[17] Kabo-Bah, A., Diji, C., Nokoe, K., Mulugetta, Y., Obeng-Ofori, D., and Akpoti, K. (2016). Multiyear Rainfall and Temperature Trends in the Volta River Basin and their Potential Impact on Hydropower Generation in Ghana. Climate, 4 (49), 1-17. https://doi.org/10.3390/cli4040049.

[18] Karanja, J. M. and Zakaria, A-R. (2018). Africa and Climate Change Refugees Quandary: Kenya Perspectives. In: Akanle O., and Adesina J. (eds) The Development of Africa. Social Indicators Research Series, vol 71. Springer, Cham. pp. 255-267.

[19] Kiros, G., Shetty, A., and Nandagiri, L. (2016). Analysis of variability and trends in rainfall over northern Ethiopia. Arabian Journal of Geosciences, 9 (451), 1-12. https://doi.org/10.1007/s12517-016-2471-1.

[20] Merabtene, T., Siddique, M., and Shanableh, A. (2016). Assessment of Seasonal and Annual Rainfall Trends and Variability in Sharjah City, UAE. Advances in Meteorology, 2016. https://doi.org/10.1155/2016/6206238.

[21] Mertz, O., Halsnæs, K., Olesen, J. E., and Rasmussen, K. (2009). Adaptation to Climate Change in Developing Countries. Environmental Management, 43 (5), 743-752. https://doi.org/10.1007/s00267-008-9259-3.

[22] Niang, I., Ruppel, O. C., Abdrabo, M. A. Essel A., Lennard C., Padgham J., and Urquhart P.., (2014). Africa. Chapter 22. In: Climate Change 2014: Impacts, Adaptation, and Vulnerability. Part B: Regional Aspects. Contribution of Working Group II to the Fifth Assessment Report of the intergovernmental Panel on Climate Change, edited by Barros, V.R., et al. (eds.)]. Cambridge, UK, and New York, USA: Cambridge University Press, pp. 1199-1265.

[23] Makate, C., Makate, M., and Mango, N. (2017). Smallholder Farmers' Perceptions on Climate Change and the Use of Sustainable Agricultural Practices in the Chinyania Triangle, South Africa. Social Sciences. 6 (30), 1-15. https://doi.org/10.3390/socsci6010030.

[24] OCED and FAO (2016). OECD-FAO Agricultural Outlook 2016-2025. Special Focus. Sub-Sahara Africa. http://www.fao.org/3/a-i5778e.pdf. Viewed on August 7, 2018.

[25] Partal, T., and Kahya, E. (2005). Trend analysis in Turkish precipitation data. Hydrol. Process. 20, 2011–2026. https://doi.org/10.1002/hyp.5993.

[26] Rahmat, S. N., Jayasuriya, N., and Bhuiyan, M. A. (2006). Precipitation trends in Victoria, Australia. Journal of Water and Climate Change, 6 (2), 278-287. https://doi.org/10.2166/wcc.2014.007.

[27] Salick, J., and Anja, B., (2007). Indigenous People and Climate Change. Missouri Botanical Garden. 1-32.

[28] Stanturf, J., Warren, M., Charnley, S., Polasky, C., Goodrick, S., Armah, F., and Nyako, Y. (2011). Ghana climate change vulnerability and adaptation assessment. Washington, DC: USAID. http://www.encapafrica.org/documents/biofor/Climate%20Change%20Assessment_Ghana_%20FINAL.pdf. Viewed on April 6, 2018.

[29] United Nations Economic Commission for Africa (UNECA) Annual Report. (2014). Thirty-third meeting of the Committee of Experts. https://www.uneca.org/sites/default/files/uploaded-documents/CoM/com2014/annual_report_2014_eng.pdf. Viewed on June 8, 2018.

[30] Yadav, R., Tripathi, S. K., Pranuthi, G., and Dubey, S. K. (2014). Trend analysis by Mann-Kendall test for precipitation and temperature for thirteen districts of Uttarakhand. Journal of Agrometeorology, 16 (2), 164–171. http://agrimetassociation.org/Images/Paper/16/4/1/229.pdf

[31] Zampaligré, N., Dossa, L. H., and Schlecht, E. (2014). Climate change and variability: Perception and adaptation strategies of pastoralists and agro-pastoralists across different zones of Burkina Faso. Regional Environmental Change, 14 (2), 769-783. https://doi.org/10.1007/s10113-013-0532-5

https://dx.doi.org/10.22161/ijeab.51.4 *ISSN: 2456-1878*

Effect of autoclaving on the nutritional quality of hard-to-cook common beans (*Phaseolus vulgaris*)

Karla A Batista[1], Wendell J Pereira[2], Bruna R Moreira[2], Cassio NS Silva[2], Kátia F Fernandes[2]

[1]Federal Institute of Education, Science and Education, Campus Goiânia Oeste, Goiás, Brazil.
[2]Polymer Chemistry Laboratory, Department of Biochemistry and Molecular Biology, Institute of Biological Sciences, ICB II, Federal University of Goiás, Brazil

Abstract— *A central composite rotatable design (CCRD) coupled to response surface methodology (RSM) was used to investigate the effect of autoclaving conditions on the antinutritional factors, functional properties, digestibility and hardness of hard-do-cook common beans. Results indicated that autoclaving conditions did not change the functional properties (water solubility, water absorption index and oil absorption capacity) of the autoclaved hard-to-cook beans. Nevertheless, the autoclaving process was able to reduce the content of trypsin (59.8%) and α-amylase (42.2%) inhibitors as well as the amount of resistant starch (45.9%), which improves the nutrient bioavailability. In addition, compared to unprocessed hard-to-cook beans, the autoclaved seed presented higher protein and starch digestibility and lower hardness, enhancing the nutritional and textural quality of the seeds. These results evidenced that autoclaving may be an interesting alternative to enable the use of hard-to-cook beans as whole grain or component in food preparations.*

Keywords— ***autoclaving, functional properties, hardness, digestibility, response surface methodology.***

I. INTRODUCTION

The common bean (Phaseolus vulgaris) is an important and inexpensive source of proteins, carbohydrate, dietary fibers, mineral and vitamins for millions of people both in developed and developing countries (Shiga, Lajolo, & Filisetti, 2004; Toledo, Rocha, Silva, & Brazaca, 2013). However, beans contain antinutritional factors that limit their protein and carbohydrate utilization.

Moreover, the storage of beans under the adverse conditions of high temperature and moisture renders them susceptible to the hardening phenomenon. This phenomenon is characterized by extended cooking time and losses in their nutritional and textural quality, which reduces consumer acceptability and the commercial value of beans (Ruiz-Ruiz, Martinez-Ayala, Drago, & González, 2008).

In this sense, the inactivation and/or removal of antinutritional components as well as changes in the textural quality are essential to improving the nutritional and organoleptic acceptability of hardened beans and turn help to effectively utilize the potential of these beans in human nutrition.

Recently, alternatives technologies have been proposed to enable the use of the hard-to-cook beans (Batista, Prudencio, & Fernandes, 2010a, 2010b, 2011; Lopes, Batista, Fernandes, & Santiago, 2012). Among the alternative technologies, the autoclaving process is a heat treatment that improves the nutritional quality of food legumes due to reduction in the antinutritional factors and increase in the bioavailability of proteins and carbohydrates (Shimelis & Rakshit, 2007).

However, there is scare information in the literature about the improvement in the nutritional quality of hard-to-cook beans as a result of autoclaving under varied conditions. In fact, the effect of the autoclaving is dependent of the parameters used in the process, such as pressure, heating temperature and autoclaving time as well as of the intrinsic properties of the material submitted to this treatment.

Therefore, the present work was undertaken to study the effect of varied autoclaving conditions on the functional properties, antinutritional factors, protein and starch digestibility and texture profile on the hard-to-cook common beans.the introduction of the paper should explain the nature of the problem, previous work, purpose, and the contribution of the paper. The contents of each section may be provided to understand easily about the paper.

II. MATERIAL AND METHODS

2.1 Materials

The seeds of common bean (Phaseolus vulgaris, c.v. BRS pontal), were provided by EMBRAPA Arroz e Feijão (Goiás, Brazil). The seeds were cleaned and stored in

polyethylene containers at 4 ºC until use. The hardening was carried out incubating the seeds in an oven at 40 ºC and 75% of moisture for 120 days (Ribeiro, Prudencio-Ribeiro, & Miyagui, 2005). All the chemical used were analytical grade.

2.2. Experimental design

The effects of autoclaving parameters in the antinutritional factors, *in vitro* digestibility, functional and texture properties were analyzed using a central composite rotatable design (CCRD) coupled to response surface methodology (RSM). The factors and levels assessed were the binomial pressure/temperature (0.5 Kgf cm^{-2}/111 ºC, 1.0 Kgf cm^{-2}/120 ºC and 1.5 Kgf cm^{-2}/127 ºC) and autoclaving time (15, 20 and 30 min) (Table 1). Samples were placed into glass flasks partially capped to allow entrance of water vapor. After autoclaving the seeds were dried out at room temperature and stored in plastic bags at 4 ºC until their use.

Table 1: Effect of autoclaving in the activity of trypsin and α-amylase inhibitors.

Run	Pressure/Temperature (X_1)	Time (X_2)	Trypsin inhibitors (UI g^{-1})	α-amylase inhibitors (UI g^{-1})
1	0.5/111	15	1025.23^{b}	5809.05^{a}
2	0.5/111	45	$977.77^{c,d}$	5534.83^{a}
3	1.5/127	15	770.75^{f}	$4337.99^{b,c,d}$
4	1.5/127	45	790.43^{f}	4002.07^{d}
5	0.5/111	30	$996.10^{b,c}$	$5295.93^{a,b,c}$
6	1.5/127	30	965.15^{d}	3766.94^{d}
7	1.0/120	15	$976.56^{c,d}$	$4211.70^{c,d}$
8	1.0/120	45	870.70^{e}	$5280.42^{a,b,c}$
9	1.0/120	30	899.42^{e}	5484.11^{a}
10	1.0/120	30	894.42^{e}	5481.11^{a}
Unprocessed hardened beans			1918.64^{a}	6512.83^{a}

Results are the mean of three determinations. Within columns, means with same superscript are not significantly different (P>0.05).

2.3. Texture analysis

The texture of the samples was evaluated through a compression test performed using a Lloyd TA1 material testing system (Lloyd Instruments Ltd, Hants, UK). Samples of unprocessed and autoclaved hard-to-cook beans were cooked at 110 ºC for 15 min and the texture tested using two flat-faced 50 mm diameter metal discs with a crosshead speed of 2 mm seg^{-1} and a compression ratio of 0.9 (i.e., a seed with an average height of 10 mm was compressed to 9 mm). The bean seeds were tested individually and the analysis employed was the return-to-start (RTS) method, measuring force under compression with a 500 N load cell. For each treatment at least 50 seeds were tested.

2.4. Antinutritional factors

The unprocessed and autoclaved seeds were ground to pass through a 0.5-mm mesh and the produced flour used to evaluate the content of antinutritional factors. The trypsin inhibitor activity was determined according to Kakade et al. (1974), using casein as the substrate for trypsin. One trypsin unit was defined as the increase of 0.1 absorbance unit at 280 nm. The unit of inhibition (UI) was defined as the relationship between the units observed in the maximum activity and the activity of the samples containing the inhibitors.

The activity of α-amylase inhibitor was determined according to methodology described by Deshpande et al. (1982), using starch as the substrate for the enzyme. One unit of α-amylase was defined to be the amount of enzyme that would produce 1.0 µmol of reducing per min of reaction. One unit of inhibition (UI) was defined as the relationship between the units observed in the maximum activity and the activity of the samples containing the inhibitors.

2.5. Functional properties

The water absorption index (WAI) and the oil absorption capacity (OAC) were determined according to methodology described by Okezie and Bello (1988). One gram of unprocessed or autoclaved hard-to-cook bean flour was mixed in 50 mL of distilled water or soybean oil and centrifuged at 3000 g for 20 min. The WAI and OAC were calculated according to the equation (1):

$$\text{WAI or OAC (g g}^{-1}) = \frac{\text{weight of wet sediment } (g)}{\text{weight of initial sample } (g)} \quad (1)$$

The water solubility (WS) analysis was carried out according to the method described by Okezie and Bello

(1988). The supernatant obtained in the WAI assay was dried to constant weight in an oven at 105 ºC. The solubility was calculated using the equation (2):

$$\text{WS (g g}^{-1}) = \frac{\text{weight of dried supernatant } (g)}{\text{weight of initial sample } (g)} \quad (2)$$

2.6. Starch quantification an *in vitro* digestibility test

The content of total starch (TS) was determined according to methodology described by Goni et al. (1997), with slight modifications. 50 mg of unprocessed or autoclaved hard-to-cook bean flour were dispersed in 5 mL of 2 mol L^{-1} KOH solution and incubated under shaking for 30 min at room temperature. After that, pH of the mixture was adjusted to 5.5 with 0.5 mol L^{-1} HCl solution and hydrolyzed using amyloglucosidase (Novozyme) for 60 min at 50 ºC. Glucose was quantified using the glucose oxidase/peroxidase reagent (DOLES) and the total starch was calculated as glucose x 0.9, after correction for the free glucose content.

In vitro starch digestibility was carried out as described by Batista et al. (2010a), with slight modifications. Samples were sequentially digested with α-amylase and amyloglucosidase and the content of released glucose was determined using the glucose oxidase/peroxidase reagent (DOLES). The extension of starch hydrolysis was calculated using equation (3):

$$\text{Starch digestibility (\%)} = \frac{(\text{content of glucose released after digestion}) \times 0.9}{\text{total starch of sample}} \times 100 \quad (3)$$

The resistant starch was calculated by the difference between total and digestible starch, according to the equation (4):

$$\text{Resistant starch (g g}^{-1} total \;\; starch) = \text{TS (total starch)} - \text{DS (digestible starch)} \quad (4)$$

The surface topography of the starch from untreated and autoclaved bean flours was observed by scanning electron microscopy (SEM). A dry, finely ground sample was placed on double-sided Scotch tape, mounted on an aluminum specimen holder, and coated with a thin film of gold under vacuum. Samples were observed under a Jeol scanning electron microscope (JSM 6610, Jeol, Japan).

2.7. *In vitro* protein digestibility test

In vitro protein digestibility was determined using the multienzymatic technique described by Hsu et al. (1977). Samples were mixed with 0.1 mol L^{-1} sodium phosphate buffer pH 8.0 and a multi-enzyme mixture (trypsin and pancreatine). The digestibility was determined by the digestion at 37 ºC, under stirring. The pH drop of the samples from pH 8.0 was recorded after 10 min of incubation. The *in vitro* digestibility was calculated according to the following regression equation:

$$\%\;\; digestibility = 425.68 - 47.64(pH \;\; drop) \quad (5)$$

2.8. Statistical analysis

The results were expressed as mean ± standard deviation (X±SD). The variance analysis (ANOVA) and Tukey's test were used to define differences in mean values of the data from 3 replicates. Results from CCRD were analyzed by using the software Statistica 7.0 (Statsoft, Inc., Tulsa, USA) and the experimental results were fitted via the response surface regression procedure, using the following second-order polynomial equation:

$$Y = \alpha_0 + \sum_i \alpha_i X_i + \sum_{ii} \alpha_{ii} X_i^2 + \sum_{ij} \alpha_{ij} X_i X_j \quad (6)$$

Where Y is the predicted response, α_0 is the intercept term; α_i is the linear coefficient; α_{ii} is the quadratic coefficient; α_{ij} is the interaction coefficients and X_i and X_j are the levels of the independent variables. The model was simplified by dropping terms that were not statistically significant ($p>0.05$) by ANOVA.

III. RESULTS AND DISCUSSION

3.1. Antinutritional factors

3.1.1. Trypsin inhibitors

Though beans are important sources of dietary protein for food nutrition, their acceptance and utilization has been limited due to the presence of antinutritional factors. The enzyme inhibitors figure among the most important antinutritional factors since they limit the hydrolysis of carbohydrates and protein, reducing the nutrient bioavailability.

Table 1 shows the effect of the autoclaving parameters in the content of enzyme inhibitors. As can be observed, the treatment by autoclaving resulted in a reduction ranging from 48.1% to 59.8% in the activity of trypsin inhibitors. Multivariate analysis showed that both pressure/temperature (X_1) and autoclaving time (X_2) significantly ($p<0.05$) affected the inhibitors activity. Fig. 1a shows the graphical representation (Pareto chart) of the magnitude of each of the parameters investigated upon inhibitors activity. As shown, the linear term of pressure

https://dx.doi.org/10.22161/ijeab.51.4 *ISSN: 2456-1878*

and quadratic and linear terms of autoclaving time negatively affected the trypsin inhibitory activity.

The graphical representation of significant effects of inter-relations and interactions of the independent variables on the content of trypsin inhibitors are depicted in Fig. 2a. As can be observed the increase of the pressure/temperature leads to a reduction in the trypsin inhibition, independently of the autoclaving time. The lower inhibitory activity was produced with autoclaving at 1.5 Kgf cm^{-2}/127 ºC for 15 min (run 3) or 45 min (run 4).

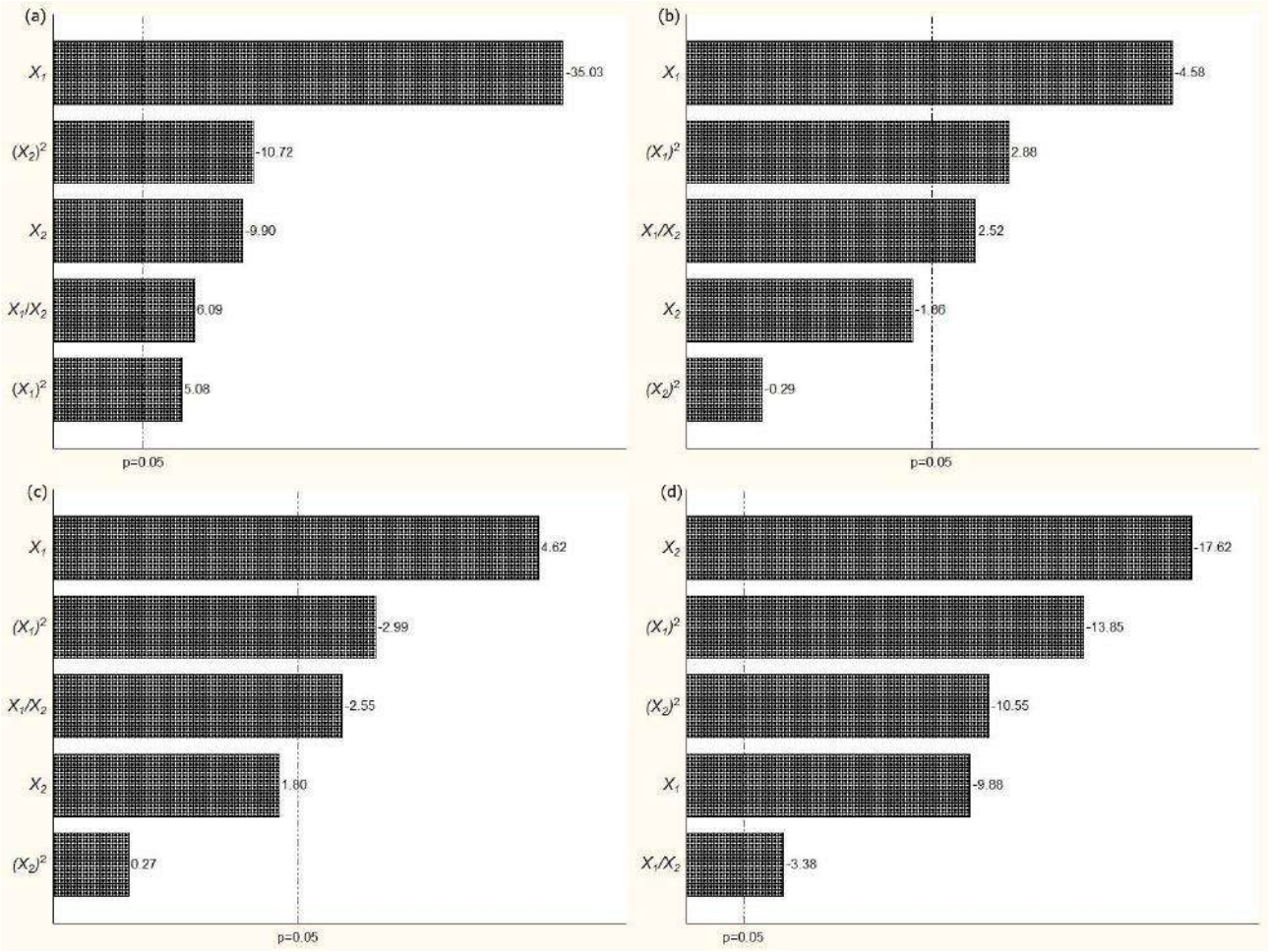


Fig.1: Pareto chart for the (a) trypsin inhibitors, (b) resistant starch, (c) starch digestibility and (d) hardness of autoclaved hardened beans.

Additionally, the regression analysis for trypsin inhibitory activity in common bean seeds showed an adequate fit of experimental values to the second-order polynomial model as a function of significant factors. The mathematical model is represented by the following equation (r^2=0.85):

$$\text{Trypsin inhibitor activity (IU g}^{-1}) = 1116.25 - 371.29X_1 + 73.28X_1^2 + 6.59X_2 - 0.17X_2^2 + 2.24X_1X_2 \quad (7)$$

Several authors reported that thermal stability of the trypsin inhibitors depends on the temperature and processing time, particles size as well as moisture content of the samples (Khattab & Arntfield, 2009; Shimelis & Rakshit, 2007). Furthermore, trypsin inhibitors are proteins that can be affected by pressure at the molecular level. Pressure may disrupt the protein non-covalent and covalent bonds leading to inactivation of the trypsin inhibitory activity (Guerrero-Beltrán, Estrada-Girón, Swanson, & Barbosa-Cánovas, 2009).

In this sense, despite the high thermal stability of some trypsin inhibitors, the combination of temperature, pressure and autoclaving time was able to reduce the inhibitory activity. This reduction in the trypsin inhibitors contributes to improve the nutritional quality of the hard-to-cook common beans submitted to autoclaving.

https://dx.doi.org/10.22161/ijeab.51.4 *ISSN: 2456-1878*

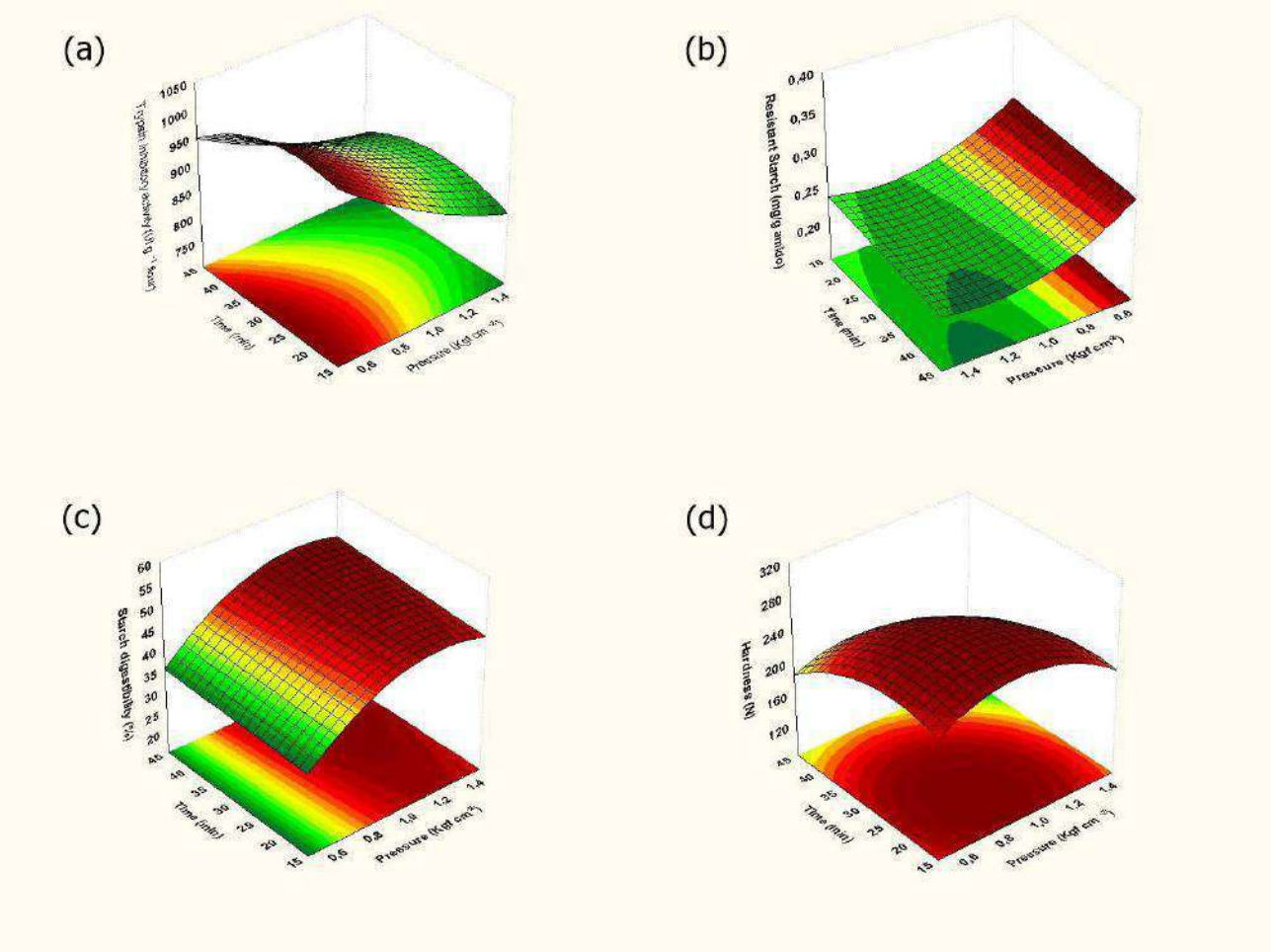


Fig.2: Response surface plot of a) trypsin inhibitors, (b) resistant starch, (c) starch digestibility and (d) hardness as a function of pressure/temperature and autoclaving time.

3.1.2. α-amylase Inhibitors

The α-amylase inhibitors are antinutritional factors responsible by inactivation of salivary and pancreatic amylases, reducing the starch digestibility and compromising the glucose bioavailability (Svensson, Fukuda, Nielsen, & Bonsager, 2004).

The effects of autoclaving on the α-amylase inhibitors activity are shown in the Table 1. As can be seen, the treatment was not able to totally eliminate the α-amylase inhibitors in the common bean seeds. The maximum reduction of α-amylase inhibitors was obtained by autoclaving hard-to-cook beans at 1.5 Kgf cm^{-2}/127 ºC for 30 min or 45 min. Results of the multivariate analysis showed that only the linear term of pressure/temperature (X_1) affected the activity of the α-amylase inhibitors in the common bean seeds. The correlation analysis showed that pressure had a negative effect (r=-0.72) in the activity of amylase inhibitors. This negative correlation indicates that as the higher the pressure, the lower the activity of the α-amylase inhibitors.

3.2. Functional Properties

The functional properties are closely related to the interactions between molecules constituting the food, which are affected by the inherent composition, structure and physicochemical properties.

Water absorption index (WAI) determines the absorption and retention of water as a function of food composition. Changes in the molecules properties after treatment, especially regarding to protein denaturation and starch fragmentation, may alter this parameter (Lopes et al., 2012).

The water solubility is other important functional property widely used as an indicator of starch degradation since increases in this index indicates an increase in the amount of soluble molecules in the material (Batista et al., 2010b; Lopes et al., 2012).

On the other hand, oil absorption is commonly associated with the presence of hydrophobic molecules such as lipids and proteins. Changes in native protein structure affect the oil absorption capacity (OAC) by

exposing hydrophobic spots, increasing the availability of sites for lipid interaction.

The effect of autoclaving conditions on the WAI, WS and OAC of the studied bean seeds is shown in Table 2. Results evidenced that autoclaving process did not change the functional properties of the hard-to-cook beans. Despite of the drastic conditions used in the autoclaving process, apparently the interaction among food components were not compromised.

Table 2: Effect of autoclaving on the functional properties of hardened common beans.

Run	Water solubility (%)	Water absorption index (g g^{-1})	Oil absorption index (g g^{-1})
1	$24.03^{a,b}$	4.44^{a}	5.37^{a}
2	16.35^{b}	4.31^{a}	5.44^{a}
3	$22.66^{a,b}$	4.19^{a}	5.46^{a}
4	$24.07^{a,b}$	5.59^{a}	5.37^{a}
5	$24.02^{a,b}$	4.42^{a}	5.12^{a}
6	$21.81^{a,b}$	4.29^{a}	5.28^{a}
7	25.97^{a}	4.32^{a}	4.80^{a}
8	$19.29^{a,b}$	4.27^{a}	5.40^{a}
9	$22.70^{a,b}$	4.82^{a}	5.32^{a}
10	$22.75^{a,b}$	4.80^{a}	5.30^{a}
Unprocessed hardened beans	$22.31^{a,b}$	4.22^{a}	5.63^{a}

Results are the mean of three determinations. Within columns, means with same superscript are not significantly different (P>0.05).

3.3. Total starch and *in vitro* digestibility

The total starch content varied from 42.6% to 44.9% in the unprocessed and autoclaved hard-to-cook bean flours. These results are in agreement with those reported in the literature for *Phaseolus vulgaris* (Rehman & Shah, 2005). The results of resistant starch and *in vitro* digestibility are shown in Table 3.

Table 3: Effect of autoclaving in the content of resistant and digestible starch of the hardened beans.

Run	Pressure/Temperature (X_1)	Time (X_2)	Resistant starch (g g^{-1} total starch)	Digestible starch (%)
1	0.5/111	15	0.35^{a}	20.34^{f}
2	0.5/111	45	0.26^{b}	40.91^{e}
3	1.5/127	15	$0.23^{c,d}$	$48.42^{b,c}$
4	1.5/127	45	$0.22^{d,e}$	$51.16^{a,b}$
5	0.5/111	30	$0.25^{b,c}$	$42.92^{d,e}$
6	1.5/127	30	$0.25^{b,c}$	$44.25^{c,d,e}$
7	1.0/120	15	0.20^{e}	54.50^{a}
8	1.0/120	45	$0.23^{c,d}$	$47.31^{b,c,d}$
9	1.0/120	30	$0.24^{b,c}$	$44.84^{c,d,e}$
10	1.0/120	30	$0.25^{b,c}$	$44.80^{c,d,e}$
Unprocessed hardened beans			0.37^{a}	15.84^{f}

Results are the mean of three determinations. Within columns, means with same superscript are not significantly different (P>0.05).

As can be seen, autoclaving process occasioned a reduction in the content of resistant starch varying from 29.7% (run 2) to 45.9% (run 7). Multivariate analysis indicates that binomial pressure/temperature (X_1) and the combined effect of pressure/temperature and autoclaving time (X_2) had a significant effect on the content of resistant starch (Fig. 1b). The response surface for the effect of autoclaving conditions in the content of resistant starch presented an overall curvilinear profile (Fig. 2b). As shown in Fig. 2b, the higher reduction in the resistant starch was found to occur in the higher pressure/temperature and autoclaving time. The minimum

https://dx.doi.org/10.22161/ijeab.51.4 *ISSN: 2456-1878*

reduction was obtained in the less drastic autoclaving condition (0.5 Kgf cm^{-2}/111 ºC; 15 min), whereas the maximum reduction in was found in the autoclaving at 1.0 Kgf cm^{-2}/120 ºC for 15 min (run 7).

The regression analysis showed an adequate fit of experimental values to the second-order polynomial model as a function of significant factors. The mathematical model is represented by the equation (8):

$$\text{Resistant starch (g g}^{-1}\text{total starch)} = 0.49\text{-}0.37X_1 + 0.12X_1^2 + 0.003X_1X_2 \quad (r^2=0.81) \quad (8)$$

The combined effect of the binomial pressure/temperature and autoclaving time probably caused changes in the morphology of the starch granule, increasing the content of molecules susceptible to α-amylase and amyloglucosidase action.

The reduction on the content of resistant starch contributed to an improvement on the starch digestibility. As can be observed in Table 3, the autoclaving process increased the starch *in vitro* digestibility that presented values varying from 20.34% (run 1) to 54.50% (run 7).

The variable pressure/temperature (X_1) and the factor interaction (X_1X_2) had a significant effect ($p<0.05$) on the starch digestibility, whereas autoclaving time (X_2) did not significantly interfere with this response (Fig. 1c). As observed in the response surface plot, higher pressure/temperature levels resulted in higher starch digestibility, particularly at higher processing time (Fig. 2c). Optimum *in vitro* starch digestibility was observed in the flour from beans autoclaved at 1.0 Kgf cm^{-2}/120 ºC for 15 min or 1.5 Kgf cm^{-2}/127 ºC for 45 min. On these conditions the starch digestibility increased more than 3-fold compared to unprocessed hard-to-cook beans.

The regression analysis showed an adequate fit of the experimental values to the second-order polynomial model as a function of significant factors. The mathematical model is represented by the following equation:

$$\text{Digestibility (\%)} = 84.26X_1 - 26.60X_1^2 - 0.59X_1X_2 - 12.82 \quad (r^2=0.82) \quad (9)$$

The digestibility degree of a starch depends on the content of molecules susceptible to hydrolytic activity, as well as on the structural characteristics of the granules. The presence of crystalline structure in the starch granules protects the glucosdic bonds against amylase activity. The autoclaving process probably caused gelatinization, modifying the crystalline structure of the starch and increasing the digestibility of the autoclaved seeds. Exception was observed only for beans autoclaved at 0.5 Kgf cm^{-2}/111 ºC by 15 min, that presented an increase only 28.4% in the starch digestibility values.

The analysis of SEM micrographs (Fig. 3) of the flour from unprocessed and autoclaved (1.5 Kgf cm^{-2}/127 ºC; 45 min) hard-to-cook beans showed that in general starch granules were round, elliptical, irregular, kidney and oval shape. They were heterogeneous in size, varying from 13 µm to 26 µm (Fig. 3a) in the unprocessed bean flours. The effect of autoclaving may be visualized by the increase in the average size of the granules, which increased from 22 µm to 53 µm (Fig. 3b).

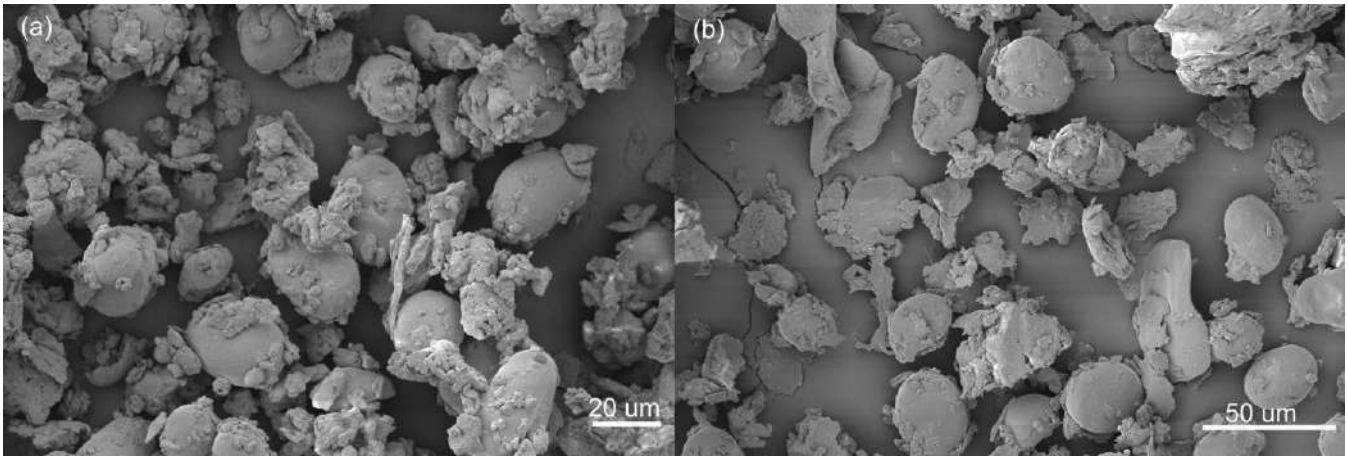


Fig.3: Scanning electron microscopy micrographs of (a) unprocessed hardened bean flour and (b) autoclaved bean flour (1.5 Kgf cm^{-2}/127 ºC; 45 min).

https://dx.doi.org/10.22161/ijeab.51.4 *ISSN: 2456-1878*

After autoclaving process, several starch granules presented a morphology resembling an amorphous mass of cohesive structure, with loss of granular appearance. This change in appearance from the regular granular shape to an amorphous structure is probably a consequence of the gelatinization occurred during the autoclaving process. The increase in the size of starch granules and the loss of original morphology is a result of swelling and subsequent gelatinization, which makes this substrate more accessible to enzymes, facilitating the starch digestibility.

Moreover, the combined effect of binomial pressure/temperature and autoclaving time caused a partial inactivation of the α-amylase inhibitors, contributing to the increase in the starch digestibility. This effect was confirmed by the correlation analysis that showed a negative correlation between the content of α-amylase inhibitors with the starch *in vitro* digestibility (r=-0.60).

3.4. *In vitro* Protein Digestibility

Results from *in vitro* digestibility of proteins are shown in Table 4. As can be observed the flours from autoclaved bean presented digestibility 41-49% higher than that of unprocessed beans. The maximum improvement in the digestibility was obtained in the autoclaving conditions of 1.0 Kgf cm^{-2}/120 ºC and 30 min (run 9 and run 10). The effect of pressure and temperature during autoclaving leads to a reduction in the content of trypsin inhibitors, improving the protein digestibility what was evidenced in the correlation analysis (r=-0.38).

In addition, the exposition of a higher number of protein linkages to enzymatic hydrolysis due to protein denaturation during the autoclaving process increases the accessibility to protein peptide bonds and thus improves the digestibility.

Table 4: In vitro protein digestibility and hardness profile of common bean seeds after autoclaving.

Run	Protein digestibility (%)	Hardness (N)
1	46.41[a]	230.99[b]
2	45.77[a]	184.14[d]
3	45.93[a]	210.56[b,c]
4	45.14[a]	135.84[e]
5	46.33[a]	230.19[b]
6	46.72[a]	199.16[c,d]
7	45.93[a]	251.72[a]
8	46.25[a]	195.43[c,d]
9	47.52[a]	257.78[a]
10	47.51[a]	257.47[a]
Unprocessed hardened beans	31.96[b]	268.07[a]

Results of digestibility are the mean of three determinations. Results of hardness are the mean of fifty determinations. Within columns, means with same superscript are not significantly different (P>0.05).

3.5. Texture analysis

Hardness is a textural problem that occurs when beans fail to soften sufficiently during the normal cooking process. The harder texture presented by hard-to-cook beans is a result of the reduction in the hydration (imbibition) and swelling of the seeds, which in turn, reduce their cookability and consumer acceptance (Nasar-Abbas et al., 2008). This reduction in the water uptake and softening of the cotyledon tissue leads to beans with inferior texture and mouthfeel. The hardness is probably caused by a poor cell separation, improper starch gelatinization and changes in the protein denaturation temperature (Yousif, Deeth, Caffin, & Lisle, 2002).

As observed in the Table 4, the unprocessed seeds had the highest values of hardness. However, the autoclaved seeds presented a reduction varying from 3.9% (run 9) to 49.3% (run 4), depending on the autoclaving parameters.

The results from multivariate analysis showed that all factors negatively affected the hardness ($p<0.05$), and the linear term of autoclaving time showed the most pronounced effect on this response (Fig. 1d). As can be seen in Fig. 2d, the hardness values decreased with improvement of the pressure/temperature and autoclaving time. The maximum reduction in the hardness was observed in the seeds autoclaved at 1.5 Kgf cm^{-2}/127 ºC for 45 min (Table 4). The regression analysis for hardness showed an adequate fit of the experimental values to the second-order polynomial model as a function of significant factors. The mathematical model can be represented by the following equation (r^2=0.91):

$$\text{Hardness}\ (N) = 56.13 + 293.59X_1 - 149.49X_1^2 + 6.54X_2 - 0.13X_2^2 - 0.93X_1X_2 \quad (10)$$

The combined effect of pressure and temperature during the autoclaving probably changed the cell adherence increasing the cell separation, which leads to seeds with lower hardness. The reduction on the hardness will contribute to improvement of the texture and palatability of the hard-to-cook beans.

IV. CONCLUSIONS

The results obtained in this study evidenced that autoclaving process improves the nutritional characteristics of hard-to-cook beans, while preserves its functional properties. The reduced activity of the enzyme inhibitors and amount of resistant starch as consequence of autoclaving, leads to an improvement on the nutrient bioavailability. Additionally, the decrease in the hardness make the autoclaved hard-to-cook beans products with better texture and palatability, compared to the unprocessed hard-to-cook seeds. These results evidenced that autoclaving may be an interesting alternative to improve the nutritional and functional quality of the hard-to-cook beans allowing its introduction in human consume as whole grain or component in food preparations.

ACKNOWLEDGEMENTS

This work was supported by CNPQ (Process numbers: 479649/2010-3 and 426284/2016-0) and CAPES (Finance code 001). We are very thankful to Empresa Brasileira de Pesquisa Agropecuária/ (EMBRAPA), for supplying the beans.

REFERENCES

[1] Batista, K A, Prudêncio, S H, & Fernandes, K F. (2010a). Changes in the biochemical and functional properties of the extruded hard-to-cook cowpea (*Vigna unguiculata* L. Walp). *International Journal of Food Science and Technology*, *45*, 794-799.

[2] Batista, K A, Prudêncio, S H, & Fernandes, K F. (2010b). Changes in the functional properties and antinutritional factors of extruded hard-to-cook common beans *Journal of Food Science*, *75*, 286-290.

[3] Batista, K A, Prudêncio, S H, & Fernandes, K F. (2011). Wheat bread enrichment with hard-to-cook bean extruded flours: nutritional and acceptance evaluation. *Journal of Food Science*, *76*, S108-S113.

[4] Deshpande, S S, Sathe, S K, Salunkhe, D K, & Comforth, D P. (1982). Effects of dehulling on phytic acid, polyphenols, and enzyme inhibitors of dry beans (*Phaseolus vulgaris* L.). *Journal of Food Science*, *47*, 1846-1850.

[5] Goni, I, Garia-Alinos, A, Manas, E, & Saura-Calixto, F. (1997). A starch hydrolysis procedure to estimate glycemic index. *Nutrition Research*, *17*, 427-437.

[6] Guerrero-Beltrán, J A, Estrada-Girón, Y, Swanson, B G, & Barbosa-Cánovas, G V. (2009). Pressure and temperature combination for inactivation of soymilk trypsin inhibitors. *Food Chemistry*, *116*, 676-679.

[7] Kakade, M L, Rackis, J L, McGhee, J E, & Puski, G. (1974). Determination of trypsin inhibitor activity of soy bean products: a collaborative analysis of an improved procedure. *Cereal Chemistry*, *51*, 376-382.

[8] Khattab, R Y, & Arntfield, S D. (2009). Nutritional quality of legume seeds as affected by some physical treatments: 2. Antinutritional factors. *LWT-Food Science and Technology*, *42*, 1113-1118.

[9] Lopes, L C M, Batista, K A, Fernandes, K F, & Santiago, R A C. (2012). Functional, biochemical and pasting properties of extruded bean (*Phaseolus vulgaris*) cotyledons. *International Journal of Food Science and Technology*, *47*, 1859-1865.

[10] Nasar-Abbas, S M, Plummer, J A, Siddique, K H M, White, P, Harris, D, & Dods, K. (2008). Cooking quality of faba bean after storage at high temperature and the role of lignins and other phenolics in bean hardening. *LWT - Food Science and Technology*, *41*, 1260-1267.

[11] Okezie, B O, & Bello, A B. (1988). Physicochemical and functional properties of winged bean flour and isolate compared with soy isolate. *Journal of Food Science*, *53*, 450-454.

[12] Rehman, Z, & Shah, W H. (2005). Thermal heat processing effects on antinutrients, protein and starch digestibility of food legumes. *Food Chemistry*, *91*, 327-331.

[13] Ribeiro, H J S S, Prudencio-Ribeiro, S H, & Miyagui, D T. (2005). Propriedades físicas e químicas de feijão comum preto, cultivar Iapar 44, após envelhecimento acelerado. *Ciência e Tecnologia de Alimentos*, *25*, 165-169.

[14] Ruiz-Ruiz, J, Martinez-Ayala, A, Drago, S, & González, R. (2008). Extrusion of a hard-to-cook bean (*Phaseolus vulgaris*, L.) and quality protein maize (*Zea mays* L.) flour blend. *LWT - Food Science and Technology*, *41*, 1799-1807.

[15] Shiga, T M, Lajolo, F M, & Filisetti, T M C C. (2004). Changes in the cell wall polysaccharides during storage and hardening of beans. *Food Chemistry*, *84*, 53-64.

[16] Shimelis, E A, & Rakshit, S K. (2007). Effect of processing on antinutrients and in vitro protein digestibility of kidney bean (*Phaseolus vulgaris* L.) varieties frown in East Africa. *Food Chemistry*, *103*, 161-172.

[17] Svensson, B, Fukuda, K, Nielsen, P K, & Bonsager, B C. (2004). Proteinaceous α-amylase inhibitors. *Biochimica et Biophysica Acta*, *1696*, 145-156.

[18] Toledo, N M V, Rocha, L C, Silva, A G, & Brazaca, S G C. (2013). Interaction and digestibility of phaseolin/polyphenol in the common bean. *Food Chemistry*, *138*, 776-780.

[19] Yousif, A M, Deeth, H C, Caffin, N A, & Lisle, A T. (2002). Effect of storage time and conditions on the hardness and cooking quality of adzuki (*Vigna angularis*). *LWT - Food Science and Technology*, *35*, 338-343.

https://dx.doi.org/10.22161/ijeab.51.5 *ISSN: 2456-1878*

Effect of Participation in Community and Social Development Project on Rural Livelihood Enhancement in North West, Nigeria

S.H Sidi, A. Hashim*, B.Z Abubakar, O.J Ladebo, A.A Uthman, F.J Yelwa

Department of Agricultural Extension and Rural Development, Usmanu Danfodiyo University, Sokoto-Nigeria.
Corresponding author: ahmadhashim70@gmail.com

Abstract**—** *The study was conducted to assess the effect of Participation in Community and Social Development on rural Livelihood enhancement in North West, Nigeria. Multistage sampling techniques were used to select CSDP beneficiaries and non-beneficiaries for the study. Data were collected from a total of 360 respondents using structured questionnaire. Data obtained was analyzed using descriptive and inferential statistics. The result showed that the age of the majority of respondents fell between 29-38 years for the beneficiaries and 39-48 years for the non-beneficiaries. Majority of the respondents were married (80.28%) from the pooled data and were male (81.11%). Approximately, 56.67% had one form of education or the other with beneficiaries more distributed in formal education. The major occupation for both beneficiaries and non-beneficiaries was farming (69.17%). With regards to CSDP participation, majority (85.6%) of the beneficiaries participated in project planning stage, 65% in project preparation stage, 71.6% participated in project implementation stage while 61.7% participated in project monitoring and evaluation stage. Participation level was rated high as majority (47.78%) of the beneficiaries participated in at least ten out of sixteen project cycles. Probit analysis showed that sex, marital status, education, monthly income and work experience were statistically related to the decision to participate in CSDP by the respondents. The double difference values was observed to be ₦92, 981.7 implying that productive assets increased more across the beneficiaries in comparison to the non-beneficiaries in the course of time. Crop farming (36.7%), cattle trading (28.3%) and livestock farming (26.9%) where the major livelihood activities of the respondents as indicated from the pooled data. Improvement in living standard, community cohesion, increased school enrolment, reduction in water borne diseases and reduction in the distance covered to school and health centers were some of the benefits beneficiaries derived from CSDP as a result of their participation. Among the major challenges facing the beneficiaries while participating in CSDP includes high cost of materials, complex protocol, payment of counterpart funds and abandoned projects. Others were lack of professional medical personnel, poor maintenance culture and possibility of elite culture. The study concludes that CSDP is promising and therefore needs to be sustained. It is therefore recommended among others that CSDP and other non-governmental organizations should encourage non-benefiting communities to participate in the project through adequate sensitization and outreaches.*

Keywords**— Social Development Project, Rural Livelihood, farming.**

I. INTRODUCTION

Approaches to development have been changing in recent years to reflect a new paradigm that emphasizes sustainability, institutional change and participatory learning process which promotes capacity building and empowerment of local people. The participation of local people in planning and managing their own development is a means of safeguarding their interest in the development process. By this, people decide their own priorities for the development and efficient use of their scarce resources which are competing for many alternative uses. They also exercise control over their own economic, social and cultural developments. Community participation in development activities was defined by Marsela (2015) as the process by which individuals, families or communities assume responsibility for their own welfare and develop a capacity to contribute to their own and the community development; it is an active process whereby beneficiaries influence the direct

and execution of development. It is regarded as one of the cornerstone for good governance. Community participation helps to enhance accountability, transparency and ensure sustainability of development initiatives.

According to Udu and Onwe (2016), over 80% of the population of developing countries resides in the rural community. For this reason, community development efforts ought to be geared towards improving the living standard of the mass of the low-income population residing in rural areas and making the process of their development self-sustaining. In support of the above statement, Oyesola, (2013) also reported that close to 80% of the population in Nigeria live in rural areas and are directly or indirectly involved in the use of land resources but majority of these rural dwellers are facing several problems, which reduces their productivity. Some of these problems include environmental constraints, infrastructural deficiencies, marketing problems, and technological constraints, institutional constraints, high cost of labour, inadequate agricultural incentive and lack of sustainable rural development programmes. This understanding, informed the community development efforts of successive governments in Nigeria targeted in the rural communities. However, most of the community development efforts failed to yield the desired results due to such factors as lack of background studies aimed at understanding the social and demographic characteristics of their target communities and groups, literacy level, pervasive poverty prevalent in those communities, hunger and disease, absence of infrastructure which improves the quality of life such as potable water, electricity and good feeder roads to mention but a few.

In view of the foregoing, this was carried and achieved the following key objectives:

i. Describe the socio-economic characteristics of CSDP beneficiaries and non-beneficiaries in the study area.
ii. Find out the levels of participation in CSDP among the beneficiaries in the study area.
iii. Determine the influence of socio-economic characteristics of beneficiaries on their participation in CSDP.
iv. Investigate the effect of CSDP participation on the livelihood assets of beneficiaries and non-beneficiaries in the study area.
v. identify the common livelihood activities of CSDP beneficiaries and non-beneficiaries in the study area
vi. Know the benefits derived by beneficiaries from CSDP participation in the study area.
vii. Investigate the major challenges to the effective participation of beneficiaries in the CSDP.

II. METHODOLOGY

The study was conducted in three States namely Katsina, Kebbi and Zamfara of North West zone, Nigeria. The North West region is made up of seven States namely Jigawa, Kaduna, Kano, Katsina, Kebbi, Sokoto and Zamfara. The North West zone is located between latitude 9^0 10 N and 13^0 50 N and longitude 3^0 35E and 9^0 00 E and it covers an area of about 102, 535 km^2(Yakubu, 2018) representing 18% of the country's total land area. The zone has a combined projected population of 52, 349, 857.67 million at 3.3% growth rate (National Population Commission, 2018).The study area has international boundaries to the north and west with Niger Republic and on the southwest with Benin Republic. The elevation of the study area is between 250 and 350 meters above sea level. Resistant crusts of laterites and ironstones characteristically cap the hills in this area. The river system represents the principal drainage network in this region (Bako, 2016).

The vegetation of the zone consists of Northern Guinea Savannah and Sudan savannah and experience low rainfall of usually less than 1000mm and the prolonged dry season (6-9 months) sustains fewer trees and shorter grasses of about 1.5-2m and few stunted trees hardly above 15m. The vegetation has undergone severe destruction in the process of clearing land for the cultivation of important economic crops such as cotton, millet, maize and wheat (Yakubu, 2011). The mean average temperature range from 18.3^0C to 28.3 ^{0}C. However, maximum daytime temperatures are for most of the year generally under 40 ^{0}C (104.0 ^{0}F) and the dryness makes the heat bearable. The warmest months are March to April when daytime temperatures can exceed 40 ^{0}C (110.0 ^{0} C). The rainy season is from May to October during which showers occur. From late October to February, during the cold season, the climate is dominated by the Harmattan wind blowing Sahara dust over the land. The dust dims the sunlight, thereby lowering temperatures significantly and also leading to the inconvenience of dust everywhere in houses (Bako, 2016).

The zone is basically an agrarian society with over 80% of the population involved in one form of animal and or crop farming or the other. They produce such crops as millet, guinea corn, maize, rice, potatoes, cassava, groundnuts, beans, wheat, sugarcane, cotton and vegetables for cash which include garlic, onions, pepper and tomatoes among others. Local crafts such as blacksmithing, weaving, dyeing,

https://dx.doi.org/10.22161/ijeab.51.5 *ISSN: 2456-1878*

carving and leather works also plays an important role in the economic life of the people. The area is also one of the fish producing areas of the country (Bako, 2016).

North Western Nigeria comprises of seven States namely Jigawa, Kaduna, Kano, Katsina, Kebbi, Sokoto and Zamfara. However, this study targeted Katsina, Kebbi, and Zamfara States. They were the States that have benefited from the activities of the CSDP. A multi-stage sampling procedure was used to select the sample for the study. The first stage was the purposive selection of the three existing senatorial zones in the selected States to ensure effective coverage and representation of communities. The second stage was the selection of one (1) Local Government Area (LGA) from each of the senatorial zones using simple random sampling technique, thus giving a total of nine LGAs. The third stage involve the selection of two benefiting communities purposively based on the presence of fully completed and functioning projects from each LGAs participating in CSDP. In addition, equal numbers of non-benefiting communities were also selected as control for estimation of counterfactual to give a total of 36 communities. The fourth and final stage involved random selection of 10 members of Community Development Associations (CDOs) from each of the 36 communities giving a total of 360 members which constituted the sample size for the study.

In order to achieve the objectives of this study both primary and secondary data were used for the study. Primary data were obtained with the aid of structured questionnaire administered in November- December, 2018 by trained enumerators. The questionnaire was tested so that the interviewers can gain familiarity with the questionnaire and provided an opportunity to apply and review the method. The focus was on assessing how respondents understand the questions and to identify any problems encountered in providing answers. Proposed changes were made and incorporated into the final questionnaire.

The instruments were used to generate information on the socio-economic characteristics of the CSDP beneficiaries and non-beneficiaries, levels of participation in CSDP, influence of socio-economic factors of the beneficiaries on their participation in CSDP, effect of CSDP on the rural livelihood assets of beneficiaries, common livelihood activities and the challenges to effective participation in CSDP. Information of the benefits derived by the beneficiaries in CSDP participation was also obtained.

The secondary data dwell on past works and reports, theses, journal articles, bulletins, newspapers and text books.

For the purpose of achieving the objectives of this research, data for this study were analyzed using both descriptive and inferential Statistics (Probit regression and Double difference Estimator). Descriptive Statistics such as frequency counts, percentages, means and standard deviations were used to achieve objectives (i), (ii), (v), (vi) and (vii) which described the socio-economic characteristics of CSDP beneficiaries and non-beneficiaries, examined the levels of participation in CSDP among the beneficiaries, examined the common livelihood activities of CSDP beneficiaries and non-beneficiaries in the study area, identified the benefits derived from CSDP by the beneficiaries and identified the major challenges to the effective participation of beneficiaries in the CSDP respectively.

Probit Regression Model was used to achieve objective iii which determined the influence of the socio-economic characteristics of beneficiaries on their participation in CSDP. A beneficiaries' decision to participate in CSDP is influenced by many socio-economic factors. The Probit model was used to analyze those factors influencing CSDP participation of beneficiaries. The decision to participate in CSDP is discrete and it takes a value of 1 if beneficiaries participate and 0 otherwise. Drawing from Von Braun and Immink (1994); Goletti (2005); Ohen *et.al*. (2013) the explicit form of the Probit model is expressed as:

$Y=\boldsymbol{\beta}_1+X_1+ \boldsymbol{\beta}_2X_2+ \boldsymbol{\beta}_3X_3+\boldsymbol{\beta}_4X_4+\ldots\ldots\ldots\ldots+\boldsymbol{\beta}_7X_7+\varepsilon i\ldots\ldots.1$

Where:

Y= Binary response defined as 1 if the respondents participates and 0 if otherwise

$\boldsymbol{\beta}$ = Estimated parameters

X1= Sex (1= male, 0= female)

X2= Age (Number of years)

X3= Marital Status (1= married, 2= single, 3= others)

X4= Educational level (Years spent in school)

X5= Household size (Number of persons in family)

X6= Monthly income (Naira)

X7= Working experience (years spent working)

$\boldsymbol{\beta}0$= intercept

ε= Error term

Double Difference Estimator was used to achieve objective iv, i.e. to determine the effect of participation in Community and Social Development Project on livelihood assets. The double difference method is a standard programme evaluation tool used to measure potential programme impact (Verner and Verner 2005). The double difference in a regression framework can be written as:

$Y_{ij}= a + DDT_it_j + \boldsymbol{\beta}T_i + t_j+ u_{ij}\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots$ (2)

https://dx.doi.org/10.22161/ijeab.51.5 *ISSN: 2456-1878*

Where:

DD= $\overline{Yi_T2017} - \overline{Yi_T2010} - \overline{Yi_C2017} - \overline{Yi_C2010}$ ..... (3)

Double difference

$\overline{Yi_T2017}$=Average livelihood assets of the beneficiaries in 2017

$\overline{Yi_T2010}$ =Average livelihood assets of the beneficiaries in 2010

$\overline{Yi_C2017}$ =Average livelihood assets of the non-beneficiaries in 2017

$\overline{Yi_C2010}$ =Average livelihood assets of the non-beneficiaries in 2010

III. RESULTS AND DISCUSSIONS

Table.1: Socio-Economic Characteristics of CSDP beneficiaries and non-beneficiaries

	Beneficiaries		**Non-beneficiaries**		**Pooled**	
Variables	Frequency	Percentage	Frequency	Percentage	Frequency	Percentage
Age						
19-28	30	16.6	53	29.4	83	23.1
29-38	61	33.8	45	25	106	29.4
39-48	53	29.7	58	32.2	111	30.8
49-58	28	15.5	22	12.2	50	13.9
59-68	6	3.3	2	1.2	8	2.2
> 69	2	1.1	0	0	2	0.6
Total	180	100	180	100	360	100
Mean	39.2		35.6		37.4	
Std. Dev.	.78.90		.73.54		.54.68	
Sex						
Male	143	79.44	149	82.78	292	81.11
Female	37	20.56	31	17.22	68	18.89
Total	180	100	180	100	360	100
Marital Status						
Single	12	6.67	30	16.67	42	11.67
Married	156	86.67	133	73.89	289	80.28
Others	12	6.66	17	9.44	29	8.06
Total	180	100	180	100	360	100
Educational Level						
Primary Education	21	11.67	25	13.89	46	12.78
Secondary Education	43	23.89	33	18.33	76	21.11
Tertiary Education	50	27.78	32	17.78	82	22.78
No Formal education	66	36.67	90	50.00	156	43.33
Total	180	100	180	100	360	100

Source: Field Survey, 2019

Table one show socio-economic characteristics of both beneficiaries and non-beneficiaries. Age was identified as the number of years at the time of interview the respondent had lived on earth. Analysis of the beneficiaries and non-beneficiaries' socioeconomic characteristics is presented in table 1 shows that 33.8% of the beneficiaries were between the ages of 29-38 years, while the same age bracket was 25% for the non-beneficiaries. The mean age of the beneficiaries was 39 years while that of the non- beneficiaries were 35 years. Therefore both the beneficiaries and non-beneficiaries

were averagely young irrespective of their status in CSDP. Although non-beneficiaries were, on average, slightly younger than their counterparts. The result implies that both beneficiaries and non-beneficiaries were of middle age and within the agricultural productive age range of 30-50 years quoted by Food and Agriculture Organization (FAO, 1997; 2005). The beneficiaries were found to be matured to make rational decisions affecting their socio-economic wellbeing in their various communities. This is in consonance with Bzugu *et.al.* (2005) who noted that younger persons participated more in agricultural and community development activities.

Community and Social Development Projects targets male and female as well as vulnerable groups of the community. The result revealed that 79.44% and 20.56% of the beneficiaries were males and females respectively, while 82.78% and 17.22% of the non-beneficiaries were males and females respectively. This implies that majority of the beneficiaries and non-beneficiaries were males which could be attributed to the current practice of purdah (women in seclusion) as the people in the area are predominantly Muslims. In Hausa culture also, men are more likely than women to participate in activities of projects like the CSDP which involve interaction with strange men. However, the finding revealed that there were more females among the beneficiaries than with the non- beneficiaries. The result was in agreement with the findings of Jonathan (2014) who found that 78.6% and 21.4% of CSDP beneficiaries were male and female respectively.

The study further revealed that 86.67% and 6.67% of the beneficiaries were married and single respectively while 73.89% and 16.67% of the non-beneficiaries were also married and single respectively. This could be attributed to the culture of the people in the area, which encourages early marriage. It could also be due to struggle to meet the needs of their families. Only 6.66% and 9.44% of the beneficiaries and non-beneficiaries had other forms of marital status such as divorced or widowed.

However, it can be readily seen that, irrespective CSDP status, majority of the beneficiaries and non-beneficiaries were married. This implies that the marital status of beneficiaries who benefited from the CSD Project did not differ markedly from those that did not benefit. This finding depicts that the beneficiaries were people that have family responsibilities which could be made easier to discharge through access to infrastructure like water, schools, health centers, etc. that are supported by CSDP. This is in line with the findings of Girei *et.al*. (2015) in their study on Impact Evaluation of Rural Health Infrastructure Sub sector of the Community and Social Development Project in Adamawa State.

On educational level, four forms of education were observed among the CSDP beneficiaries and non-beneficiaries, these were primary, secondary, tertiary or no formal education. Findings from the study in table 2 further show that 23.89% and 18.33% of the beneficiaries and non- beneficiaries had secondary education respectively. Also 27.78% of the beneficiaries had tertiary education while only 17.78% of the non- beneficiaries had tertiary education. These results shows that rural people in the study area actually valued education and it further confirms that the beneficiaries were sufficiently enlightened so as to appreciate the importance of involvement and participation in community project delivery. Also the result conforms to the studies of Fawole and Tijani, (2012) and Adesida and Akunola, (2015) that high literacy level can enhance participation and better understanding of any initiative programme. However, 36.67% and 50% of the beneficiaries and non-beneficiaries stood as those without formal education respectively. Non- formal education in this research consisted of adult literacy and Qur'anic education.

Result of the study shows that 42.2% and 56.11% of the beneficiaries and non-beneficiaries had household size of 1-5 persons respectively. According to the results 31.7% and 23.34% of the beneficiaries and non-beneficiaries had household size of 6 – 10 persons respectively while only 5.6% of the beneficiaries had more than 20 members. The mean household size was about 8 for the beneficiaries of CSDP and about 6 for the non-beneficiaries. This shows that CSDP beneficiaries have relatively large household size than the non-beneficiaries and it may not be unconnected to the common practice of polygamy and extended family systems in the study area. This agrees with Thomas *et.al.* (2018) findings that the average household size of market participants was 8 people.

The occupational distribution of the beneficiaries and non-beneficiaries shows that they had five primary occupations. They were farmers, traders, public servants, and artisan and agro processors.

https://dx.doi.org/10.22161/ijeab.51.5 *ISSN: 2456-1878*

Table 2: Distribution of beneficiaries and non-beneficiaries according to socio-economic characteristics 2

	Beneficiaries		Non-beneficiaries		Pooled	
Variables	**Frequency**	**Percentage**	**Frequency**	**Percentage**	**Frequency**	**Percentage**
Primary Occupation						
Farming	115	63.89	134	74.44	249	69.17
Trading	20	11.11	16	8.89	36	10.00
Public service	34	18.89	19	10.56	53	14.72
Artisan	8	4.44	7	3.89	15	4.17
Agro Processing	2	1.11	4	2.2	6	1.67
Unemployment	1	0.56	0	0	1	0.28
Total	**180**	**100**	**180**	**100**	**360**	**100**
Monthly income						
< ₦10,000	19	10.6	29	16.1	48	13.3
₦10,000-₦50,000	133	73.9	139	77.2	272	75.6
₦51,000-₦90,000	17	9.4	12	6.7	29	8.0
₦91,000-₦130,000	6	3.3	0	0	6	1.7
₦131,000-₦170,000	3	1.7	0	0	3	0.8
>₦170,000	2	1.1	0	0	2	0.6
Total	**180**	**100**	**180**	**100**	**360**	**100**
Mean	₦34000		₦24775		₦29458	
Std. Dev.	2454.63		1259.94		1399.63	

Source: Field survey, 2019

Table 2 shows that majority of the beneficiaries and non-beneficiaries were into farming as primary occupation with non-beneficiaries of CSDP being more distributed within the category than their counterparts. However, among those reported on the other categories (Trading, Public service, and artisan), the beneficiaries were proportionally higher than their counterparts.

The result in table 2 shows that 63.89% of the beneficiaries of CSDP and 74.44% of the non-beneficiaries were into farming while 18.89% and 10.56% of beneficiaries and non-beneficiaries respectively were public servant. Some 11.11% and 8.89% of beneficiaries and non-beneficiaries were into trading and only few were artisan. The findings disagree with Aderinoye-Abdul wahab *et.al.* (2015) who reported that the major occupation for income generation in communities was trading on non-farm produce (39.8%). Also the fact that most of the beneficiaries were farmers means that they are based in rural areas where there is serious lack of functional infrastructure such as roads, schools, hospitals etc. This lack of infrastructure might have motivated them to seek the assistance of the CSDP in providing some of these much needed infrastructure.

Result in Table 2 showed that majority of the beneficiaries (73.9%) had a monthly income of between ₦10,000 - ₦50,000 slightly below the non-beneficiaries with 77.2%. About 9.4% and 6.7% of the beneficiaries and non-beneficiaries had a monthly income of between ₦51,000-₦90,000 respectively, while 10.6% and 16.1% of the beneficiaries and non-beneficiaries had less than ₦10,000 monthly income respectively. Very few (1.1%) of the beneficiaries had a monthly income of ₦171,000 and above. The mean income for the beneficiaries was ₦34,141.67 while non-beneficiaries were ₦24,775. The result supports the findings of Okereke-Ejiogu *et.al.* (2015) who found the mean monthly income of participants to be ₦38,268.52. This implies that the beneficiaries earn some money at the end of the month and this could encourage their participation in community development projects like CSDP as they can afford to pay the levies if such need arises.

https://dx.doi.org/10.22161/ijeab.51.5 *ISSN: 2456-1878*

Table 3: Levels of Participation of Beneficiaries in Community and Social Development Project

Project Planning stage			
Identification of projects	43	23.9	4th
Project selection	50	27.8	3rd
Need assessment	61	33.9	1st
Project preparation stage			
Consultation with technical/professionals	20	11.1	14th
Decision on the scope of micro-project	19	16.7	7th
Preparation of community development plan	15	8.3	16th
Counter fund contribution	52	28.9	2nd
Project Implementation stage			
Community meetings	35	19.4	5th
CPMC training	15	8.3	15th
Labour contribution at project site	30	16.7	8th
Selection of project sites	26	14.4	11th
Procurement of materials	23	12.8	12th
Monitoring and Evaluation stage			
Serving in project committees	33	18.3	6th
Problem solving	27	15	10th
Reporting and consultation	30	16.7	9th
Writing of physical progress report	21	11.7	13th
Total	*500		

Source: Field Survey, 2018

***Multiple responses**

Community participation is very important tool for developmental process in any country. It was observed that CSDP beneficiaries in the study area participated in the sixteen basic stages of the CSDP project cycle. Table 3 shows the distribution of beneficiaries according to the stages of CSDP project cycle they were engaged in. The revealed that the beneficiaries participated more in project planning stage, project implementation stage than in project preparation and monitoring and evaluation stages. Results showed that majority (85.6%) of the beneficiaries participated in project planning stage with 23.9% participated in project identification, 27.8% in project selection and 33.9% in project need assessment. The high participation in the Project Planning Stage could be attributed to sensitization and awareness creation carried out by the Community and Social Development Project agencies in the study area and also the Participatory Rural Appraisal method employed in assessing the needs of the communities. Planning stage takes into consideration the interest of the different segments of the communities (men, women, youth, elderly and vulnerable persons) not just at the implementation stage hence the highest participation.

It was also observed from the result that 65% of the beneficiaries were involved in project preparation stage, of which 28.9% participated in counterpart contribution, 16.7% in the decision on the scope of micro project, 11.1% participated in consultation with technical or professionals while 8.3% were involved in preparation of community development plan. The study shows that beneficiaries' participation in the project preparation stage had lesser participation than the project planning stage. The only component of the project preparation stage that had high percentage (28.9%) and ranked second of beneficiaries 'participation was the community counterpart contribution. Responses during the interview sessions revealed more of participation of CPMC members at this stage than the generality of the community members. Consultation with technical persons/professionals was said to be the responsibility of the CPMC members who were meant to report back to the community members during community general meetings as a form of feedback mechanism.

In project implementation stage, a total of 71.6% of the beneficiaries were involved out of which 19.4% participated in community meetings, 14.4% participated in the selection of project site, 16.7% participated in labour contribution at project site, 12.8% in procurement of project materials and only 8.3% were participated in community project management committee training. This revealed that the communities are responsible for financial management, procurement and other implementation aspects of the

projects, and they are only supported by the state agency (SA), LGA and other relevant experts where the communities deem it necessary. Monitoring is concerned with the continuous and routine measures enshrined to ensure that activities required for successful completion are adopted and followed. Labour contribution was ranked 8th and second component. In the context of this study, levels of participation of beneficiaries in Community and Social Development Project in the study area fall into three categories, namely: Low, Medium and High based on the frequency of participation in different stages of participation. Table 3 reveals that 16.67% of the beneficiaries had low participation having involved in less than 5 levels of activities. Majority of the CSDP beneficiaries (47.78%) had high participation having participated in more than 10 out of 16 levels of activities while 35.56% had medium participation having involved 6-10 levels of CSDP activities.

Table 4: Distribution of beneficiaries based on their levels of participation in community and social development project

Levels of participation	Frequency	Percentage
<5 (Low)	30	16.67
5-10 (Medium	64	35.56
>10 (High)	86	47.78
Total	**180**	**100**

Source: Field survey, 2019.

Socio-economic factors influencing beneficiaries' participation in community and social development project

Probit analysis was conducted to determine the influence of socio-economic characteristics of beneficiaries and non-beneficiaries on their participation in CSDP. The result is presented in Table 4. The ratio statistics indicated by chi-square statistics are highly significant (p< 0.0000). This suggests that the model has a strong explanatory power. The pseudo R^2 is 0.0686 meaning that the regressors were able to explain 69% of CSDP participation in the study area. It was observed that out of seven independent variables considered for analysis, five were significant. The significant factors included the sex, marital status, level of education, monthly income and work experience.

Sex was positively and significantly related to the decision to participate in CSDP by beneficiaries at 1% level of probability; this implies that respondents who were male are more likely to participate in CSDP in the study area compared to women. This observation is consistent with the findings of Abdul-Hanan and Anang (2018) and Thomas *et.al.* (2018). the reason for this finding is that in a typical rural setting, household heads are usually males who are the decision-makers in terms of access to resources and participation in programmes. Women often need the permission of their husbands to participate in programmes thus constraining their participation rates. The hypothesis is therefore rejected for this variable. Marital status was significantly related to the decision to participate in CSDP by beneficiaries at 5% level of probability; this implies that beneficiaries that have family responsibilities are more likely to participate in CSDP than other respondents.

The result also showed that level of education had a negative coefficient (-.1952248) and significant at 10 percent level of probability. It should be recalled that a negative sign on the coefficient implies that as level of education increases, perceived level of participation of CSDP decreases. Similarly, a positive sign indicates that with a unit increase in a particular variable there is also an increase in the perceived level of participation in CSDP within the study area. This implies that the higher the level of education of the respondents, the less the probability of participation in CSDP activities. Education decrease of Participation correlate with the report by Sani (2018) said there could be cases that educated households have the high chance of engaging themselves in other non-farm related activities such as sideline business, involvement in the administration that leave them with little time to participate in community development activities. The result is in conformity with findings of Adeyemo and Kayode (2012) who found that education (r=-2.641; P<0.00) has significant but negative coefficient with level of sustainability of community projects within the study area.

Monthly income was a significant factor influencing participation in the CSDP programme. This implies that people with relatively higher income are more likely to participate in CSDP in the rural areas. The reason might be, those with low income are very much busy looking for what to eat and therefore may not necessarily have time to partake in the activities of CSDP.

The probit model results show that working experience was significantly associated with the probability of CSDP participation. This shows that experienced people were more likely to participate in CSDP relative to unexperienced ones. Our result here is plausible and expected. More experienced house heads have overtime, developed some understanding of programmes that can help to improve their socio-economic wellbeing. The result is in agreement with the findings of Udo, (2014) who underlined that working experience among other factors have influence in programme participation in Nigeria.

However, age and household size was inversely related to participation since the value of their coefficient was found to be (.002832 and .006075) and was not statistically significant (0.856 and 0.870) at either 1% or 5% level of probability. It is therefore shows that the age and household size of the respondents have no influence on participation in the CSDP activities. It was hypothesized that beneficiaries' socio-economic factors have no influence on CSDP participation. The finding showed that sex with z value of (0.007), marital status (0.024), education (0.064), monthly income (0.024) and work experience (0.044) had significantly influenced beneficiaries' participation at 1%, 5%, 10%, 5% and 5% level of probability respectively. It is therefore concluded that socio-economic factors have influence on beneficiaries' participation in the CSDP; hence, the null hypothesis is hereby rejected.

Table 5: Socio-economic factors influencing participation in Community and Social Development Project (CSDP)

Variables	Coefficient	Standard Error	z- Value
Sex	0.9265	0.3432	0.007***
Age	0.0028	0.0173	0.870ns
Marital status	-0.3907	0.1731	0.024**
Education	-0.1952	0.1054	0.064*
Household size	0.0060	0.0334	0.856ns
Monthly income	0.0000	5.34e-06	0.024**
Work Experience	0.0353	0.0175	0.044**
Constant	-0.9608	0.6499	0.139
Log likelihood	0-232.4		
Pseudo R^2	0.0686		
Prob> chi^2	0.0000		

***, **, * significant at 1%, 5% and 10% probability respectively.

Distribution of beneficiaries based on the challenges experienced in CSDP participation

Objective iv was to identify the challenges experienced by the beneficiaries in CSDP participation. As revealed in table 5, majority of the beneficiaries (52.2%) complained that high cost of materials was their major challenge during the implementation of the project. The CPMC were given the mandate to award contract and source materials locally based on the budget approved by the CSDP agency. The price of the materials were most of the time go high as against the approved unit price. Next in ranking is the challenge of complex protocol as reported by 48.3% of the beneficiaries as participating communities have to undergo series of protocol before partaking into the programme. The result is in consonance with the findings of Adeyemo *et.al.* (2014) who stated some protocol the community undergo, that community members have to be mobilized and sensitized, groups have to be formed and legally registered, group officers have to be elected and bank account have to be opened if not already in place. Additionally, Participatory Rural Appraisal have to be conducted for need assessment, Local Development Plans have to be drawn, submitted and approved. Counterpart fund of at least 10% also have to be paid before possible disbursement of funds for project implementation. These listed conditions requires significant time and therefore seen as a challenge by most beneficiaries. Inability of the beneficiaries to contribute to the levies placed on them towards the provision of project counterpart fund and other important developmental activities was another challenge reported by 38.9% of the beneficiaries. The result are in tandem with the findings of Adejoh (2015) who reported financial constraints as challenge affecting women participation in CSDP in Kogi State. About 38.3% of the beneficiaries reported slow decision making process as a challenge facing communities regarding CSDP. The community have to draw community development plan

(CDP). The CDP is a comprehensive community plan for development activities within a community and contains a portfolio of micro-projects. Once the SA approves a CDP, the micro projects would be implemented one after the other in accordance with approved plan. Thus, unless the first micro project selected for implementation is successfully completed, grants shall not be released by the SA for the others. The CDP will then be submitted to LGDO who the recommend to LGRC for approval. This takes time and delay the approval.

The study also identified other factors such as abandoned project and possibility of elite capture as challenges faced by communities regarding CSDP as reported by 35.6% and 22.8% of the beneficiaries respectively. Some project were abandoned due to financial constraints and sometimes washed away by rains in the case of road project. The elite who acted as a threat to hijacked community project capitalize on the perceived weaknesses of some community members to pay certain fees and thereafter act as lords over them. Poor maintenance culture as reported by about 23.9% of the beneficiaries was seen as a problem being faced by communities. The beneficiaries explained that active participation diminishes immediately after project implementation. Even though committees are set up at various stages to ensure the sustainability of the project, community members are not so cooperative in that regard. The levies charged for the maintenance of the project are not paid. Finally, lack of qualified medical personnel was reported by 17.8% of the beneficiaries as a challenge, as most medical personnel deployed to CSDP clinics are not professionally qualified to attend to serious issues of health concern.

Table 6: Distribution of respondents based on the challenges

Challenges	Frequency	Percentage	Rank
High cost of materials	94	52.2	1st
Complex Protocol	87	48.3	2nd
Payment of counterpart funds	70	38.9	3rd
Slow decision making process	69	38.3	4th
Abandoned project	64	35.6	5th
Poor maintenance culture	43	23.9	6th
Possibility of elite capture	41	22.8	7th
Lack of qualified medical personnel	32	17.8	8th
Total	*500		

Source: Field survey, 2018

Multiple responses

IV. CONCLUSION AND RECOMMENDATIONS

The combined influence of socio-economic variables (Sex, marital status, level of education, monthly income and work experience) have made positive and significant contributions to beneficiaries' participation in CSDP activities at 1%, 5% and 10% level of probability. However, age and household size of the beneficiaries were not significant. The result revealed that the beneficiaries participated more in project planning stage, project implementation stage than in project preparation and monitoring and evaluation stages. Beneficiaries were also found to have high participation in CSDP activities. High cost of materials, complex protocol and payment of counterpart funds were identified as the major challenges to the effective participation in CSDP. It is recommended that CSDP and non-governmental organizations should encourage non-participating communities to participate through adequate sensitization and outreaches. Female operation officers and facilitators should be recruited in the future for project of this nature. This would enhance greater participation of women in the project. Communities should expedite actions in the payment of counterpart fund so as to attract many more projects in the community. Disbursement of funds for the project should be timely to avoid unnecessary rise in the price of working materials.

REFERENCES

[1] Abdul-Hanan, A., & Anang, B.T., (2018). Factors Influencing Participation of Cocoa Farmers in the Government Spraying Programme in Ghana. *Asian Journal of Agricultural Extension, Economics and Sociology, 22(2);* 1-9.

[2] Adejoh, S.O. (2015). *Participation of Rural Women in Community and Social Development Project and its*

Contribution to their Livelihood in Kogi State, Nigeria. (Unpublished Ph.D Thesis). Ahmadu Bello University, Zaria.

[3] Adekunle, A.A., Oluwa, T.A., & Ladele, A.A. (2005). *Bridging the Communication Gap between Scientists and Farmers in Katsina State of Nigeria*. International Institute of Tropical Agriculture, Ibadan, Nigeria. 7pp.

[4] Aderinoye-Abdulwahab, S.A., Nwachukwu, S.C., Salawu, O.L., & Popoola, P.O., (2015). Assessment of Livelihood activities of Rural Farmers in Kwara State, Nigeria. *Ethiopian Journal of Environmental Studies and Management, 8(2);* 120-129.

[5] Adeyemo, P. A., Oladosu, I. O., Kayode, A. O. and Orimafo, P. K. (2014). Level of participation of community-driven development approach beneficiaries of World Bank assisted projects in South Western Nigeria. *Journal of Humanities and Social Science19*(11), 64-70.

[6] Akinwalere, B.O and Ajibola, M.O (2015). Assessment of Rural Infrastructural Development Projects in Ondo State: Case Study of Ondo State Community and Social Development Agency. *American Journal of Experimental Agriculture10 (2)* pp. 2.

[7] Anelle, D (2012). *A Brief Note on the Condition of Rural Areas in Nigeria*. Retrieved from: www.vanguardngr.com

[8] Bako, M.M. (2016). *Analysis of adoption of improved rice production technologies by rice farmers in North-Western Nigeria*. Unpublished PhD thesis, Department of Agricultural Extension and Rural Development, Usmanu Danfodiyo University, Sokoto.

[9] Girei, A.A., Dire, B., & Haruna, U. (2015). Impact Evaluation of Rural health Infrastructure Sub sector of the Community and Social Development Project, Adamawa State, Nigeria. *Scholar Journal of Agriculture and Veterinary Sciences, 2(2B)* 138-143.

[10] Goletti, F. (2005). *Agricultural commercialization, value chains and poverty reduction. Making Markets World Better for the Poor Discussion Paper No.7*. Hanoi: Asian Development Bank.

[11] Hidayat, U. T. (2011). *The effectiveness of Community-based Development in Poverty Reduction: A Descriptive Analysis of a Women Managed NGO in Rural Pakistan.* Tokyo.: Institute of Economic Research, Hitotsubashi University.

[12] Jonathan, O.E. ((2014)). Empirical Evidences of the Impact of Community Education and Social Development Projects on Rural Bayelsa, Nigeria. *British Journal of Education 2 (3)*, Pp 65-74.

[13] Marsela, N.M., (2015). *The importance of Community Participation in Development Project at Local Level: A case of Dodoma Municipal Council.* Unpublished PhD Dissertation, Department of Local Government and Management, Mzumbe University, Tanzania. 1-6pp.

[14] Odishika, V. A and Adedeji, O. (2016). Community Driven Development (CDD) and Rural poverty Alleviation in Nigeria: A Bottom-up Development Approach. *Sociology and Anthropology 4 (6)*, 439.

[15] Ohen, S. B., Etuk, E. A. and Onoja, J. A. (2013). Analysis of Market Participation by Rice Farmers in Southern Nigeria, *Journal of Economics and Sustainable Development, 4(7);* 6-11.

[16] Okereke-Ejiogu, E. A. (2015). Assessment of Households' Participation in Community and Social Development Project (CSDP): A case study of Imo State, Nigeria. *Advances in Research Journal 5 (2)*, 1-9.

[17] Oyesola, O.B. (2013) Rural Dwellers perception on Effect of Infrastructural Facilities on Livelihood activities in Akinyele Local Government Area of Oyo State, Nigeria. *Journal of economics and rural development, 16* (1), 83.

[18] Sani, H. (2018). Influence of socio-economic factors influencing farmers' Participation in State agricultural development Programme in Zamfara State, Nigeria. *International Journal of Research in Applied, Natural and Social Sciences.* 6 (8) 11-22.

[19] Sylvia, S., (2014). *Determinant of Participation in Livelihood Diversification among Peasant farmers in Rural Uganda.* Unpublished MSc Dissertation, Department of Population Studies, Makerere University, Ugaanda. 22p.

[20] Thomas B. Y , Aymar Y. B, Bio M. T , Jean-Louis F, Dapola E. C., Yacouba Y., Georges S., Fourvouon S. and Mariam M. D. (2018). Socio-Economic Factors Influencing Small-Scale Farmers' Market Participation: Case of Rice Producers in Dano. *Sustainability, 10*(4354), 6-11

[21] Udo N.C., (2014). Socio-economic Factors Influencing Farmers' Participation in Community- Based Programme in Abia and Cross River States of Nigeria. *J. Agr. Ext*. 18(1):48-61.

[22] Udu, L.E., and Onwe, S.O (2016). Approaches to Community Development in Nigeria, Issues and Challenges: A Study of Ebonyi State Community and Social Development Agency (EB-CSDA) *Journal of Sustainable Development*. 9(1) 296-307.

[23] Umar, B.F. (2013). *Impact Evaluation of Zamfara State Community and Social Development Project.* Gusau, Nigeria: ZCSDP.

[24] Von Braun, J. and Immink, M. D. C. (1994). *Nontraditional vegetable crop and food security among smallholder farmers in Guatemala*. John Hopkin university press, Baltmore, MD. 189-203.

[25] Yakubu, H. D. (2011). *Socio-economic Factors Affecting the Adoption of ICTs by Extension Workers in North- West Zone of Nigeria*: Unpublished M.Sc Dissertation, Department of Agricultural Extension and Rural Development. Usmanu Danfodiyo University, Sokoto.: Sokoto

https://dx.doi.org/10.22161/ijeab.51.6 *ISSN: 2456-1878*

Role of Local Non-Governmental Organizations (NGOs) in Community Development in Zamfara State, Nigeria

A Hashim, S.H Sidi, B.Z Abubakar, B.F Umar, H.M Aliero, F.J Yelwa

Department of Agricultural extension and rural development, Usmanu Danfodiyo University, Sokoto, Nigeria.
Email: Corresponding author: ahmadhashim70@gmail.com

Abstract— *The study was conducted in Zamfara State, Nigeria to assess the role of local Non-Governmental Organizations (NGOs) in community development. A questionnaire was administered to 200 randomly selected beneficiaries of NGOs activities. Another questionnaire was administered to 58 purposively sampled officials of local NGOs for the study. Descriptive such as (frequencies and percentages) and inferential statistics like t-test and correlation were used to analyze the data collected from the field. The study revealed that, local NGOs contributes significantly to community development in various ways such as through community self-help efforts; training and re-training of community members and awareness creation and sensitization of community members. It was revealed that, these efforts resulted to many improvements such as increased enrolments of children in schools; improved health care service delivery; and increased agricultural yields. The research has shown that, beneficiaries provide water; labour and venue during execution of community development efforts. The study identified problems militating against smooth running of NGOs activities: inadequate funds and personnel; low level of beneficiaries 'commitments; and low level of government support. It was recommended that, government, individuals and development partners should provide more financial, technical and human resources support to NGOs in order to make them effective and efficient in delivery of community development services.*

Keywords— ***Role; Local Non-Governmental Organizations (NGOs); Community development.***

I. INTRODUCTION

Poor performance of governments in meeting the socio-economic quests of its citizens has been identified as one of the reasons behind the proliferation of Community Based Organizations (CBOs) and Non-Governmental Organizations (NGOs) in the present Millennium. Non-Governmental Organizations (NGOs) are voluntary clubs, societies and associations or groups of people within the society or community that come together to achieve certain objectives (United Nations 1990). They may have similar identities and share some needs and aspirations. These organizations are non-profit or business oriented. Non-Governmental Organizations (NGOs) are organizations or societies that have little or no direct bearing with any of the three levels of government; in most cases they do not have permanent paid officials (United Nations, 1990).

There is increasing evidence that all three sectors of society-government, private and civil society (NGOs)-have important roles to play in nations' building and particularly in development efforts to improve the quality of lives of citizens. When they are able to work well together they mutually reinforce each other's work and can together do what none of them can do on its own. These collaborative and working relationships have not been documented in the case of Zamfara State.

The study was conducted to achieve the following Objectives:

i. Describe the socio-economic characteristics of beneficiaries of community development activities;
ii. Identify the roles played by the NGOs in community development;
iii. Find out Roles of beneficiaries towards execution of community development activities in the area;
iv. Identify the perception of beneficiaries towards NGOs in community development;
v. Find out forms of assistance provided by government in supplementing the efforts of the NGOs in the study area; and

https://dx.doi.org/10.22161/ijeab.51.6 *ISSN: 2456-1878*

vi. Investigate problems of NGOs as perceived by the beneficiaries in the study area;

II. METHODOLOGY

The research was conducted in Zamfara State, Nigeria; Gusau is the capital of the state. The area is located on latitude 12^0 10'N and longitude 12^0 16'E. It was created in October 1996 by the then head of state Late General Sani Abacha; It has 14 Local government areas sparsely distributed across the state. It was created from former Sokoto state. The State is bordered in the north by Niger republic, to the south by Kaduna State, in the east it is bordered by Katsina state and the west by Sokoto, Kebbi and Niger States, (Zamfara State government Bulletin, 2012). The total area covered by the state is 39,762km2 with of 3,602,356 people (NPC, 2006).Ninety percent of populations are farmers and or offering services to the growing agricultural industries in the area (NPC, 2006). The climate of the area favors growth and development of millet, sorghum, maize, cotton, groundnuts, cowpea, tobacco and locust beans etc. and also is generally relative to its tropical position. Highest maximum and minimum annual temperatures are 38^0c and 18^0c respectively. A hamattan period, which falls between December to February, is characterized by very cold temperature and dust-laden winds, often accompanied by thick fog of high intensity. The main annual rainfall is 853.40mm, which is enough to support healthy growth of lot of tropical crops (Zamfara State Government Bulletin, 2012).The soil is generally sandy loam moderately fertile and easy to cultivate. There are two major ethnic groups; Hausa and Fulani who were predominantly farmers, rearers and businessmen, other tribes are also available which include; Yoruba, Igbo, Ibira, Tiv, Nupe, Zabarmawa and many other minority tribes. Islam is the predominant religion in the area. The vegetation characteristics are of the Sudan Savannah type with abundant grassland and sparse trees. The area is situated in the southern Sudan Savannah zone. (Zamfara State Government Bulletin, 2012).

The research work was carried out using a list of 58 registered local NGOs operating in the state. This formed the study sample frame. The list was obtained from Zamfara State Agency for NGOs affairs. In selecting the beneficiaries, purposive and simple random sampling techniques were used. Village heads and 9 other persons from each village were selected from 20 villages. A total of 200 copies of beneficiaries' questionnaire were administered. In each of 20 surveyed villages, a Focused Group Discussion was held.

Both primary and secondary data were used. Well-structured questionnaires were framed and relevant information from Non-Governmental Organizations (NGOs) and selected community members who benefited from NGOs interventions were obtained. The questionnaires were administered to officials of the NGOs and beneficiaries in the study area. A Focused Group Discussion guide was also developed and used. The data collection was done in May 2018 using the three instruments highlighted above. Other techniques used in data collection included; likert scale, triangulation and ranking.

III. RESULTS AND DISCUSSION

Table 1: Socio-Economic Characteristics of Beneficiaries of NGOs efforts (n=200)

Parameters	Frequency	Percentage
Sex		
Male	141	70.5
Female	59	29.5
Age Group (Years)		
10-20	40	20.0
21-30	56	28.0
31-40	77	38.5
41-50	22	11.0
51-60	5	2.5
Education status		
Islamic education	26	13.0
Adult education	15	7.5
Primary education	99	49.5
Secondary education	39	19.5
Tertiary education	11	5.5
Occupation		
Civil servant	48	24.0
Farmer	114	57.0
Trader	26	13.0
Other petty business	12	6.0
Marital status		
Married	170	85.0
Single	17	8.5
Divorced	11	5.5
Widow	2	1.0
Family size (Persons)		
Less than 5	99	49.5
5-10	86	43.0
11-20	13	6.5
21 and above	2	1.0

https://dx.doi.org/10.22161/ijeab.51.6 *ISSN: 2456-1878*

Family Income Level per Annum			
N100,000 – N500,000		111	55.5
N500,001 – N1,000,000		79	39.5
N1,000,001 above	and	10	5.0
Total		**200**	**100.0**

Source: Field Work 2018

Table 1 represents socio-economic characteristics of beneficiaries; sex distribution shows that, 70.5% of the beneficiaries were male while only 29.5 were females. This implies that, males benefits more from the community development initiatives of NGOs perhaps due to cultural norms that placed women folk as second class citizens. In northern Nigeria, women participate less in decision making and community development especially self-help efforts for reasons of long patriarchy. This is in line with Yusuf (2002) who reported that women in northern Nigeria are constrained to engage in crop production by several factors such as religious and traditional restrictions also women are exposed to time consuming domestic work or household activities. According to him, women spend long hours on collection of water from distant sources, collection of firewood, processing cooking food. Age distribution of beneficiaries shows 38.5% falls within the age range of 31-40years. While, only 2.5% were within the age range of 51-60 years. This implies that, youth participate more in community development may be due to their activeness and physical structure. Youthful age is the most critical age for human productivity in the present millennium; old age might not be able to contribute as youth can. The age bracket fallen within the age bracket defined by David *et al. (2009)* as economically productive in a population that is 15-65 years; also the age range is an incentive for a long lasting development. Educational qualification shows that, 49.5% of the beneficiaries had primary education while the least percentages of (7.5) and (5.5) were recorded by adult and tertiary education respectively. Education is a weapon for transformation and making people release their full potentials. This result is not a surprise considering myths and misconceptions around western education in the study area. In agreement to this finding, Aliyu (2001) documented that, low level of western education in Northern Nigeria is due to holding firmly to the ethics of Islam and having reservation against western style of education. Similarly, Edet (2004) and Cohen (1987) established that, education is a key ingredient of political, economic and social empowerment which invariably could affect engagement in rural development programmes. It has clearly show, active participation in community development efforts is dependent upon level of education of an individual or group of individuals. The more educated a community is, the more enlighten and the more they actively partake in community development in their area. Occupation distribution shows that 57% of the beneficiaries have farming as their main occupation while the least (6.0%) were engaged in petty businesses as such handcraft, Kiosk keeping, barbing saloon, commercial motorcycles riders and water vendors. This is not unexpected because the agricultural nature of the area justified the need for emancipating the resource poor farming communities through community development activities. This is in line with Yusuf (2002) who reported that, Zamfara State is cited slightly sloppy topography with good soil suitable for agricultural production. This makes the area to have a slogan "Farming is our Pride" this finding proves that, communities participating in agricultural activities like, land clearing, irrigation and processing in the study area are practically possible. The marital status indicates 85% of the beneficiaries are married while only 15 are either single, divorced or widows. This means that marriage is not a barrier to participation in community development activities. The married are more matured and responsive to development issues. This is in agreement with findings of Sani, (2008) who argued that; marriage is highly cherished high in the rural communities of the study area. The beneficiaries' family sizes shows that, 49.5% of the beneficiaries have family size of less than 5 while the least (1.0%) have a family size of 20 and above. This implies that, majority of the beneficiaries have less or equals to a maximum of 5 family members; they have lesser burden that might hinder their participation in community development activities. However, the family size could have effect on the income level of the beneficiaries as some of the members could contribute to the economic status of the entire family. This is contrary to findings of Manga (2012) who reported that, the average household size was 10 people per household. According to her, this large house hold size is a common characteristic of rural household especially in Northern Nigeria where polygamy is mostly practiced. The income level reveals that, 55.5% of the beneficiaries have an income of N100, 000 – N500, 000 per annum while the least 5% have an income level of N1, 000,000 and above per annum. This is not a

surprise because better economic status can enhance participation in community development activities through increased financial contributions towards execution of community development activities. This could also be related to kinds of occupations of the beneficiaries. If an individual is gainfully employed, he or she stands better chance of generating more incomes than unemployed. The tendency to make cash contributions for community development efforts is much higher in the former than the later.

The study revealed a positive correction (0.631) between family income and occupation. Meaning, there is significant relation between occupations and family income levels of beneficiaries. Beneficiaries with low incomes were found to be farmers and low grade civil servants. Traders and business persons reported to have higher income levels, may be due to high profits being generated from their business. To further buttress the point that, income is related to occupation of an individual and in turn can affect participation to community development activities.

Table 2: Distribution of roles of NGOs in community development as identified by NGOs

Roles of NGOs	Frequency	Percentage
Promoting community self-help efforts	20	34.5
Training and re-training of beneficiaries	44	75.9
Promoting active Participation	28	48.3
Provision of Welfare Package	17	29.3
Awareness Creation and Sensitization	33	56.1
Enhance Income Generation activity	14	24.1
Improving Social Services	23	39.7
Mentoring Community Based Organizations	16	27.6
Representation of citizens at decision making	21	36.2

Source: Field Work 2018

Table 2 shows that, 75.9% of the NGOs had attributed immense roles they played through training and re-training of community members to appreciate the value of self-help efforts. This was vigorously pursued through enlightenment and sensitization for self – awakening. This is in line with Anyanwu, (1990) that, Citizens' participation could best be achieved through training and re-training on the values of self-help which is of the people, by the people and for the people. Also Aliyu, (2012) reported that, training and capacity building is aimed at strengthening the effectiveness of an individual, which in turn enhances the level of participation in any community development programme. The finding is similar to that of Aliyu, (2012) quoting Oshuntogun (1996) who found that training strengthens people and makes them to be very efficient in discharge of their activities. Only 24.1% played role of promoting income generation activities perhaps due to their nature, most of the Local NGOs are not for profit. This implies that, community members might not be economically independent of local NGOs near future.

Table 3: Distribution of beneficiaries based on their perceptions of the contribution of NGOs in community development.

Contribution/effects of NGOs activities	Frequency	Percentage
Increased enrolment of children in schools	154	77
Enhanced health care service delivery	141	70.5
Increased yield and production levels	133	66.5
Increased women participation in development	97	48.5
Increased income levels of community	77	38.5
Increased supply of basic social services	65	32.5
Improved shelter and clean environment	48	24
Supported eradication of extreme poverty	39	19.5
Increased access to information and knowledge	30	15
Reduced maternal and child mortality rates	12	6

Source: Field Work 2018

Table 3 shows a number of effects of community development activities executed in the area by the NGOs. Increased enrolment of children in schools through mobilization and enlightenment took the lead. Where people are educated their capacities would be developed and could facilitate sense of belonging and ownership of community

https://dx.doi.org/10.22161/ijeab.51.6 *ISSN: 2456-1878*

development activities and partake in community development programme. This is in line with Samuel (2005) who reported that, education being a weapon for human transformation had resulted into meaningful achievements of the yearnings and aspiration of the community through organized effort that enable the beneficiaries to realize and exploit their potentialities to the fullest. The least score recorded for reduction of maternal and child mortality is not unconnected with the fact that, majority of local NGOs don't have interest on maternal and child health related issues; the international development organizations takes lead of promoting maternal and child health related issues in the state.

Table 4: Comparism of roles played by NGOs and beneficiaries perception about NGOs contributions in community development.

Parameters	**N**	**Mean**	**Std. Deviation**	**Df**	**Level of Significance**	**$t_{Calculated}$**	**$t_{Critical}$**	**Remarks**
Roles played by NGOs in community development	9	11.122	4.4480					
				17	0.05	0.446	1.740	Not significant
Beneficiaries perception towards contributions of NGOs	10	10.000	6.2498					

Source: **Field work, 2018** p<0.05

Table 4 shows that, $t_{Calculated}$ and $t_{Critical}$ values are 0.446 and 1.740 respectively at 0.05 level of significance. Since $t_{Calculated}$ = 0.446 < $t_{Critical}$ = 1.740, this shows that, there is no significant difference in roles and perception of the contributions of NGOs in community development in the study area. Therefore, both NGOs and beneficiaries indicated same roles and contributions of NGOs to community development. This implies that, community members recognize the roles played by the local NGOs in promoting community development in their areas and that, the local NGOs claims have been verified.

Table 5: Distribution of beneficiaries based on their rules in community development

Types of roles	**Frequency**	**Percentage**
Provision of communal labour	188	94
Provision of water	149	74.5
Provision of venue for training activities	125	62.5
Provision of transportation	97	48.5
Making cash donations	60	30
Active participation include planning	37	18.5
Others	22	11

Source: Field Work 2018

Table 5 shows that, beneficiaries have participated actively in various community development activities through provision of labour and other forms of assistance that are essential in the realization of community development Objectives. This is not a surprise because community members are the greatest resources for their own betterment. This is contrary to the findings of Samuel (2005) that reported monetary contribution and family labour were the main modes of community participation by both male and female (40.3% and 37.5%) respectively. According to him, majority of female group (47.9) quoting Akinsorotan and Olujide (2003) who assessed the level of participation of community development association members in self-help project in Lagos State where 52% of the respondents contributed money, time and labour, 14% participated by providing money, only 4% and 26% served as implementation committee members.

https://dx.doi.org/10.22161/ijeab.51.6 *ISSN: 2456-1878*

Table 6: Perception of beneficiaries on NGOs efforts towards community development (n=200)

Parameters	Frequency	Percentage
Improvement of living standards		
SA	108	54.0
A	66	33.0
N	14	7.0
D	9	4.5
SD	3	1.5
Promotion of self-help Efforts		
SA	59	29.5
A	89	44.5
N	34	17.0
D	12	6.0
SD	6	3.0
Improve Welfare		
SA	81	40.5
A	66	33.0
N	29	14.5
D	11	5.5
SD	13	6.5
Extent of contribution		
Great	75	37.5
Moderate	88	44.0
Low	30	15.0
None of All	7	3.5
Total	**200**	**100.0**

Source: Field Work 2018

Key: SA = Strongly Agreed; A =Agreed; N = Neutral; D = Disagreed; SD = Strongly Disagreed

Table 6 shows that, the NGOs had contributed immensely towards improvement of lives and expectancy and had resulted in the improvement of community welfare and productivity. This is likely so because people always appreciates who benefit them. The indifferent could be attributed to low level of education of the beneficiaries that could not critically analyze previous and current situations to be able to tell if there is any change. In agreement to this finding is the result of Manga (2012) who reported that support is sometimes extended to individuals, group and communities and is aimed at improving their livelihood activities and is expected to result in their improved wellbeing.

Table 6 shows that, beneficiaries had attested to the fact that, NGOs promotes self-help efforts in the community. This is not surprising as government and non-governmental interventions are not permanent in the communities and hence, the need for the community member to help themselves. This is in line with Zaki (2003) 'rural communities are faced with numerous problems and government alone cannot provide everything for all its citizens, hence community groups are formed to arrest such problems. This was further corroborated with principles of community development by Anyanwu (1999) who reported that *principle of self-help* – this is the main end product of community development. Enables people to exploit to their advantages the resources which could otherwise be dormant. Make use of under-utilized labour. Increase the component and confidence of a community in the heading of its affairs. Constitute a pre-requisite for survival in the modern world. Enables people to change the way they look at their responsibilities, and help them to cultivate the sense of local initiative and effort. Enhances the development of democratic values and processes; promote the idea of ultimate control by the people; fosters a substantial degree of freedom by individual and groups; leads to a considerable amount of government decentralization. Promotes wide spread citizens participation.

Table 6 shows, that beneficiaries have confessed that, NGOs activities in the study area are enormous to changing welfare status of beneficiaries through series of welfare packages and income generation activities that improves economic capabilities of beneficiaries. If people sufferings are eliminated they tend to be removed from bondage of poverty and insecurity. Their lives will be better up. This result is similar that of Adeola, *et al.* (2008) that conducted a research on the effect of the federal government special programme on rice yield and farmers' income and reported an increase in mean income of the respondents after the programme and concluded that the programme has impacted positively on the respondents.

Table 6 shows that NGOs efforts in improving community are of great importance to the beneficiaries and had acknowledged NGOs roles. These efforts were well received and appreciated by the beneficiaries. The least score 3.5% that reported none at all might be due to lack of awareness that a particular effort is done by an NGO not government.

Table 7: Comparison of the perception of the roles of NGOs by the beneficiaries according to Sex.

Sex	N	Mean	Std. Deviation	Df	Level of Significance	$t_{Calculated}$	$t_{Critical}$	Remarks
Male	141	7.50	2.609	198	0.05	0.695	1.645	Not significant
Female	59	7.81	3.442					

Source: **Field work, 2018**

Table 7 revealed that, $t_{Calculated}$ and$t_{Critical}$ values are 0.695 and 1.645 respectively at 0.05 level of significance. Since $t_{Calculated}$ = 0.695 < $t_{Critical}$ =1.645. This indicates that, there is no significant difference in perceptions of the roles of NGOs by beneficiaries based on sex. Hence, both males and females in this study perceived the roles of NGOs as the same. Even though women were not many in the study but the result shows same perceptions perhaps men had influenced their choice. Women are always submissive to the will of their husband, hence the influence.

Table 8: Distribution of beneficiaries' perception of problem of NGOs

Problems of community Development	Frequency	Percentage
Financial constraints	189	94.5
Inadequate personnel to implement program	164	82
Low level of community commitments	120	60
Low level of government support to NGOs	123	61.5
Negative attitudes of beneficiaries to community development	20	10

S**ource: Field Work 2018**

****Multiple response***

Table 8 shows that NGOs face a number of problems that hinder their development. Inadequate funds and funding support constitutes a great challenge towards achieving their desired objectives. While the least was the poor attitudes of beneficiaries to community development. This is in line with Keck and Kathryn, (1998)" Funds play a vital role in any NGO for execution of its projects, Programmes or activities, which are development oriented to their community. Hence funding agencies, donors, sponsors, are very important to all NGOs".

Table 9: Distribution of problems of NGOs as perceived by themselves (NGOs)

Problems of NGOs	Frequency	Percentage
Financial constraints	50	86.2
Inadequate personnel/staff	24	41.4
Low level of community commitments	21	36.2
Low level of government and donor support	29	50
Negative attitude of beneficiaries to community development	29	50

S**ource: Field Work 2018**

****Multiple response***

Table 9 shows that, NGOs have realized that they face some problems that affect their full performance and financial constraint took the lead. The least 10.1% was the poor attitudes of beneficiaries towards community development activities. This implies that, inadequate funds and funding support constitutes a great challenge towards achieving the desired objectives. This means that, achieving significant impacts of promoting community development initiatives in the study area might not be realized if funds are not available.

Table 10: Comparison of proportion of NGOs problems by beneficiaries and NGOs

Person square Chi-	Number of valid cases	Df	Level of Significance	$x^2_{Calculated}$	$x^2_{Critical}$	Remarks
Beneficiaries and NGOs	200	4	0.05	7.134	9.49	Not significant

Source: **Field work, 2018**

https://dx.doi.org/10.22161/ijeab.51.6 *ISSN: 2456-1878*

Table 10 shows that $x^2_{Calculated}$ and $x^2_{Critical}$ values are 7.134 respectively at 0.05 level of significance. Since $x^2_{Calculated}$ = 7.134 < $x^2_{Critical}$ = 9.49. This indicates that, there is no significant difference in the proportion of problems of NGOs as perceived by the beneficiaries and the NGOs themselves. Hence, both have same perceptions in regards to Problems that hinder NGOs progress towards community development programs. This implies that, NGOs and beneficiaries can jointly work together to address the impediments identified and that, the problems identified is no longer that of NGOs alone but the communities themselves.

Table 11: Distribution of beneficiaries according to forms of assistance required from government to supplement the efforts of the NGOs

Forms of assistance	Frequency	Percentage
Increase funding support	149	74.5
Increase level playing ground	126	63
Collaboration and engagement	130	65
Support program implementation	130	65
Administrative support	87	43.5
Capacity development opportunities	35	17.5
Enabling environment for donors	33	16.5

Source: Field Work 2018

Multiple response

Table 11 shows that, for NGOs to be effective in their functions, there is need for government and other institutions to provide assistance such as funds and skills to the NGOs. In line with Samuel (2005) who reported that, government provides funds, contracts and training opportunities to give special encouragement to NGOs activities in priority areas without undermining NGOs autonomy and independence; broad agreement is sought with NGOs on such priorities by establishing formal consultation with NGO leaders. For a such as the Council for Advancement of People Action and Rural Technology (the body which channels government funds to NGOs in India) and the forthcoming Community Action Program (a local government scheme for financing NGOs and community initiatives in Uganda) are illustrations.

IV. CONCLUSION

From the findings of the study, it could be concluded that NGOs has immensely contributed to community development initiatives through training and re-training as well as sensitization and mobilization of beneficiaries on the importance of self-help. It could be concluded that beneficiaries also contributes to their development if adequately mobilize to do so. It was further more concluded that, beneficiaries have positive perception to the critical roles of NGOs towards improving their livelihoods. It could be also concluded that, occupation of beneficiaries determines their income level and that is no significant difference in the perceptions of beneficiaries on the roles and problems of NGOs in the study area.

V. RECOMMENDATIONS

After careful examining the roles of local NGOs in community development in the study area and results that depicts challenges to community development in the area; the following recommendations were proposed to make NGOs effective in delivering community development services.

1. Government, individuals and other agencies should increase their support to NGOs. these support should include, provision of enabling working environment, provision financial, materials and technical report.
2. Beneficiaries of community development programmes and activities should continue to support activities of the NGOs in their areas and should ensure maintenance and sustainability of the completed community development projects.
3. Citizens should form and more local NGOs to complement government efforts towards community development.
4. NGOs should be more dedicated in execution of community development activities especially through promotion of self-help. They should also be interested in promoting livelihoods of rural dwellers;
5. NGOs should always carry beneficiaries along planning, execution and even evaluation of any community development activities in their domain. This will promote sustainability and ownership of the project.
6. Beneficiaries should increase their participation levels in community development through actively contributing to all community development efforts.

Maintenance culture of beneficiaries for project executed by the NGOs should be sustain;

REFERENCES

[1] Adeola, R.G, O.O Adebayo and G.O Oyelere (2008), Effects of Federal Government Special Rice Programme on Rice Yield and Farmers income in Oyo state. *International Journal of Agricultural Economics and Rural Development.* Vol. 1 No. 1 pp 124.

[2] Aliyu, B (2002), Introduction to Sociology Studies. 1st ed. Kano; Zonnet Press. Pp 47

[3] Anyanwu, C.N. (1999), *Introduction to Community Development*. Gabesther Education Publishers Ibadan, Pg 143.

[4] Cohen, J.M (187). *Rural Development participation. Concepts and measure for project design implementation and evaluation*. 3rd ed. New York: itace Publishers, Pp 123

[5] David, H.D.S, Abdulrahman, R.M, Sani, S. Kushawa and M Nasiru (2009), Resource use efficiency in irrigated Crop Production by Fadama users in Bauchi State, Nigeria; implication for Food Security and Poverty alleviation. Proceedings of the 23rd annual conference of Farm management of Nigeria, Faculty of Agriculture, Usmanu Danfodiyo University, Sokoto, Nigeria, 14th – 17th December, 2009, Pp 111.

[6] Edet, C (2004). *Impact of rural development on infrastructural development. A case study of Zaria LGA* An unpublished thesis, ABU Zaria Pp. 187

[7] Keck, M.E and S Kathryn (1998), *Non-governmental organizations and their efforts.* Oxford University Press; pp 284

[8] Manga, A.T (2012), Role of International Fund for Agricultural Development/Community based Agriculture and Rural Development in Poverty Reduction among women in Kebbi state, Nigeria. Unpublished MSc. Thesis Department of Agricultural Extension and Rural Development, Usmanu Danfodiyo University Sokoto, Nigeria

[9] National Population Commission NPC) 2006 census data. www.npc.gov.ng

[10] Samuel, U.T (2005),the Role of Non-governmental Organizations (NGOs) in Development. A paper prepared for Coalition of NGOs in Delta State. Nigeria Village square Publishers.

[11] Sani, M.K (2008). Impact of Community based Agricultural and Rural Development Programme on Livelihoods of selected LGAs of Sokoto State, Nigeria, Unpublished Msc thesis. Department of Agricultural Economics and Extension, Usmanu Danfodiyo University Sokoto, Nigeria.

[12] United Nations (1990), Community New York, Oxford University Press.

[13] Yusuf, A. (2002), Roles of Government in Financing NGOs. The Central Bank of Nigeria Journal of Economics 39(5); Pp 162

[14] Zaki, B.A (2003), *Rural Community development*. Lecture manuscripts, Usmanu Danfodiyo University, Sokoto; Unpublished document.

[15] Zamfara State Government Bulletin, 2012 First edition, State Government Press. Pp. 45.

https://dx.doi.org/10.22161/ijeab.51.7 *ISSN: 2456-1878*

Preparation and Evaluation of Goat Manure-Based Vermicompost for Organic Garlic Production in Manyatta sub-county, Kenya

Gichaba, V. M[1*]; Ndukhu, H. O[2]; Muraya, M[3]; Odilla, G. A[4]; Ogolla, F. O[5]

[1]Department of Plant Sciences, Chuka University, P.O. Box 109-60400, Chuka Kenya
Email: vincent.gichaba@gmail.com, Mobile: 0729212384
[2]Department of Plant Sciences, Chuka University, P.O. Box 109-60400, Chuka Kenya
Email: hndukhu@gmail.com, Mobile: 0727485052
[3]Department of Plant Sciences, Chuka University, P.O. Box 109-60400, Chuka Kenya
Email: moses.muraya@gmail.com, Mobile: 0704999099
[4]Department of Agricultural Economics, Agribusiness Management and Agricultural Education, Chuka University, P.O. Box 109-60400, Chuka Kenya
Email: gilbura@yahoo.com, Mobile: 0725679586
[5]Department of Biological Sciences, Chuka University, P.O. Box 109-60400, Chuka Kenya
Email: ogolla.fredy@gmail.com, Mobile: 0708576198

Abstract— *Application of vermicompost in crop production results in improved soil chemical properties. Studies relating to use of vermicompost as alternative to synthetic fertilizers have gotten considerable attention as the demand for organically produced agricultural products increases. Goat manure has been reported to be rich in nitrogen, phosphorous and potassium. However, preparation and utilization of goat manure-based vermicompost in organic garlic production in the study area is scanty. Thus, farmers have solely relied on chemical fertilizers in garlic production which is a health and environmental hazards and causes ground and surface water pollution due to nitrate leaching. In Manyatta sub-county of Eastern Kenya, farmers use synthetic fertilizers in garlic production which is not sustainable despite having readily available goat manure which can be composted for use. The aim of this study was to prepare and evaluate effectiveness of goat manure-based vermicompost for organic garlic production in Manyatta sub-county, Kenya. Goat manure-based vermicompost was prepared at KALRO Embu station, Embu County between July–November 2018. The vermicompost obtained was dried, screened and filled into bags and was used for growing garlic. A sample of goat manure-based vermicompost was analysed for chemical properties and the results showed that it had very high total N (1.79%), very high available P (52 ppm), very high exchangeable K (1.75 Cmol Kg^{-1}) and it was moderately alkaline (pH 7.73). Hence, goat manure-based vermicompost is recommended in the organic production of garlic in Manyatta sub-county of Eastern Kenya.*

Keywords— ***Goat manure, organic production of garlic, vermicompost.***

I. INTRODUCTION

Vermicompost is a complex mixture of earthworm faeces, humified organic matter and microorganisms, which when added to the soil or plant growing media, increases germination, growth, flowering, fruit production and accelerates the development of a wide range of plant species (Lazcano and Dominguez, 2011). Vermicompost greatly increases microbial activity and nutrient availability. In addition, it contains most nutrients in plant available forms such as nitrates, phosphates, and exchangeable calcium and soluble potassium which are responsible for increased plant growth and crop yield (Arancon *et al.*, 2004). Vermicompost makes soil structure spongy, improves bulk and real density, porosity, increases aggregate's stability and soil structure, and increases the rate of water penetration in the soil and aeration (Ahmadabadi *et al.*, 2011).

Although there have been many studies relating to the benefits of using vermicompost as a fertilizer source (Alsina *et al.*, 2013), there is no available research which has focused on use of goat manure in preparation of vermicompost and on the optimum application rates of vermicompost on garlic in the study area. Mikile (2001) conducted a study which showed that goat manure is one of the richest sources of nutrients that can be used for soil enrichment. The study showed that, in terms of nutrients

composition of different manures, cattle manure had the lowest nitrogen, phosphorus and potassium contents followed by sheep manure and goat manure had the highest content of nitrogen, phosphorous and potassium.

A wide range of organic residues, including sewage sludge, animal wastes, crop residues and industrial refuse are increasingly being converted by earthworms to form vermicompost (Pascal *et al.*, 2010). Earthworms breakdown the organic residues, which stimulate greater microbial activity, increase nutrient mineralization rates, and rapidly convert the wastes into a humus-like substance that has a finer structure than ordinary composts while possessing greater and more diverse microbial population (Atiyeh *et al*., 2000). Earthworms have an important influence on soil structure, forming aggregate and improving the physical conditions for plant growth and nutrient uptake. During vermicomposting, earthworms eat, grind, and digest organic wastes with the help of aerobic and some anaerobic micro flora, converting them into a much finer, humified, and microbial active material (Degwale, 2016). The organic carbon in vermicompost releases the nutrients slowly and steadily into the soil and enables the plant to absorb the available nutrients (Lalitha *et al.*, 2000).

Vermicompost has been reported to contain plant growth regulators and other growth influencing materials produced by micro-organisms which can contribute positively towards better crop yields (Atiyeh *et. al*., 2002). Studies have indicated effect of vermicompost on an improvement in soil physical properties, soil fertility and uptake of mineral nutrient (Azarmi *et al*., 2008). However, the effective preparation of goat manure-based vermicompost on garlic production is not yet fully understood in Kenyan condition. Therefore, the present research focused on studying the effective way of making goat manure-based vermicompost so as to enhance the soil chemical properties, growth and yield of garlic in the study area.

II. MATERIALS AND METHODS

2.1 Study site

The study was conducted at Kenya Agricultural and Livestock Research Organization station in Embu. The site lies at a latitude of 0.4762°S and longitude 37.4702°E. Manyatta sub-county is located on the eastern slope of Mount Kenya in Embu County. According to Jaetzold and Schmidt (1983), Embu County lies in the lower midland 3, 4 and 5 (LM3, LM4 and LM5), upper midlands 1, 2, 3 and 4 (UM1, UM2, UM3 and UM4) and inner lowland 5 (IL 5) at an altitude of approximately 500 m to 1800 m above sea level (a.s.l.). It has an annual mean temperature ranging from 17.4 to 24.5°C and an average annual rainfall of 450 mm to 1400 mm. The rainfall is bimodal with long rains falling from around March to June and short rains from around October to December. It has *humic nitisols* soils. The prime cropping activity is maize intercropped with beans though livestock keeping is also equally dominant. Various agricultural activities have been carried out in the region hence the rationale behind its selection (Kisaka *et al.*, 2015).

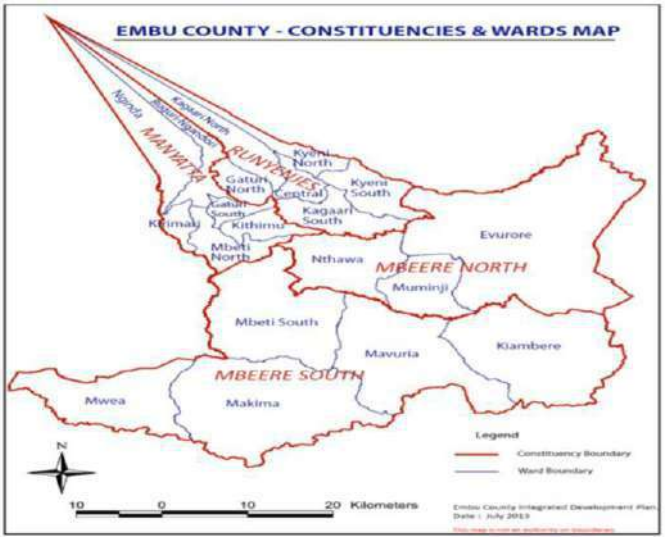


Fig.1: Map of Embu County (Sourced from Kenya mpya County maps, 2012)

2.2 Preparation of Goat Manure-Based Vermicompost

Goat manure was used as the main raw material to prepare vermicompost. A vermicompost bed was constructed using bricks and mortar on the walls. The bed measured three metres long by one-metre-wide by one-metre-high according to Ansari and Sukhraj (2010). A shed was erected above the vermicomposting bed to prevent direct sunlight and rain and also fenced around using wire chain link to prevent predators of earthworms like chicken and other birds.

A basal layer composed of broken bricks followed by a layer of coarse sand to the thickness of seven centimetres was placed inside this bed to ensure proper drainage according to Ramnarain *et al.* (2019) and restrict earthworm movement towards the soil layer. A layer of 15 cm of loamy soil was placed at the top and moistened. Small lumps of fresh goat manure were scattered over the soil. This acted as an active growing medium for earthworms according to Rekha *et al.* (2018).

Two thousand and eight hundred red wiggler earthworm (*Eisenia fetida)* species were introduced to facilitate decomposition of the materials according to Mbithi *et al.* (2015). The earthworms were sourced from AAA growers in Naromoru, Laikipia County. This was followed by ten

https://dx.doi.org/10.22161/ijeab.51.7 *ISSN: 2456-1878*

centimetres thick layer of dry grass, dry banana leaves and dry bean husks which were placed above it to act as bedding material of the worms. Dry goat manure weighing 100 kg was placed and spread on these materials in the bed up to ten centimetres thick. The same set of layers was continued till a height of one metre according to Rekha *et al.* (2018).

This was followed by sprinkling uniformly five litres of water to the vermicompost bed in order to keep it moist and facilitate easy earthworm movement in these materials. Water also prevents desiccation of earthworms. Gunny bags were placed on the top to cover the materials.

The vermicompost bed was kept moist by sprinkling two litres of water once a week and this continued up to the seventh week. Turning of these materials was done once after 15 days and it was done gently to avoid injuring earthworms since they have soft bodies. Goat manure-based vermicompost was harvested after 120 days according to Ramnarain *et al.* (2019). This was after the earthworms were found sticking to the under surface of gunny bags indicating that composting process was complete.

The goat manure-based vermicompost from the bed was harvested and spread on a polythene sheet. From this harvested goat manure-based vermicompost, adult worms and young ones were handpicked and isolated. The goat manure-based vermicompost obtained was dried under a shed for one day, screened and was filled into bags ready for organic growing of garlic.

A sample of goat manure-based vermicompost was analyzed for pH using a digital pH meter (Jones, 2001), total N using kjeldahl method (Bremner and Mulvaney, 1982), available P using extraction with 0.5 M $NaHCO_3$ according to methods of Olsen *et al.* (1954) and exchangeable K using Flame photometer (Jackson, 1967). The analysis was done at the University of Nairobi, upper Kabete campus soil laboratories.

2.3 Goat manure based-vermicompost and goat manure sample analysis results

A sample of goat manure-based vermicompost (GMBV) and goat manure (GM) used in the experiment were analysed for chemical properties and the results are presented in Table 1.

Table 1: Chemical analysis of goat manure-based vermicompost and goat manure samples used in the experiment

Type of manure	Parameters	Characterization	Very high	High	Medium	Low	Very low
GMBV	pH-H_2O (1:2.5)	7.73			>75.5-7.0	<5.5	
	TN (%)	1.79		>0.7	0.5-0.7	<0.5	
	P (ppm)	52	>46	26-45	16-25	10-15	<9
	K (Cmol Kg^{-1})	1.75		>0.60	0.31-0.60	0.21-0.30	
GM	pH-H_2O (1:2.5)	8.0			>75.5-7.0	<5.5	
	TN (%)	0.32		>0.7	0.5-0.7	<0.5	
	P (ppm)	24	>46	26-45	16-25	10-15	<9
	K (Cmol Kg^{-1})	0.59		>0.60	0.31-0.60	0.21-0.30	

Legend: GMBV – Goat manure-based vermicompost, GM – Goat manure, TN – Total Nitrogen, P – Phosphorous, K – Potassium

The results of chemical analysis of goat manure-based vermicompost used in the study showed that it had very high total nitrogen, very high available phosphorous and exchangeable potassium and it was moderately alkaline (Table 1) according to the ratings based on the ranges given by Hazelton and Murphy (2007). For goat manure which was also used in the study, the results showed that it had high total nitrogen, medium available phosphorous and exchangeable potassium and it was moderately alkaline (Table 1). The ratings were based on the ranges given by Hazelton and Murphy (2007). Hence, these results showed that goat manure-based vermicompost had higher nutrient availability (nitrogen, phosphorous and potassium) than goat manure that was used in this experiment.

2.4 Initial Soil Characteristics

The soil samples of the experimental site were analysed for soil chemical properties before planting and the results are presented in Table 2.

https://dx.doi.org/10.22161/ijeab.51.7 *ISSN: 2456-1878*

Table 2: Initial soil characterization of the study site

Soil parameter at Manyatta study site		Comparison soil nutrient rating scale				
Parameters	Soil characterization	Very high	High	Medium	Low	Very low
pH-H_2O (1:2.5)	6.33			>75.5-7.0	<5.5	
TN (%)	0.03		>0.7	0.5-0.7	<0.5	
P (ppm)	4.57	>46	26-45	16-25	10-15	<9
K (Cmol Kg^{-1})	0.26		>0.60	0.31-0.60	0.21-0.30	

Legend: TN – Total Nitrogen, P – Phosphorous, K – Potassium

The results showed that the pH of the soil at Embu was 6.33 which is moderately acidic based on the ranges given by Hazelton and Murphy (2007). This value lies within the ranges of 6.0 and 7.0 which is considered as optimum soil pH suitable for garlic (DAFF, 2017).

The initial total nitrogen content (0.03%) of the soils at Embu was very low when compared to Tadesse (2015) classification. According to Tadesse (2015), nitrogen content of soil of less than 0.05% is rated very low, between 0.15 – 0.25% is rated medium and greater than 0.25% is rated high. The deficiency of N results in reduced onion yields with respect to size and weight of the bulb (Mohammad and Moazzam, 2012). The initial low N level in the study area may be attributed to lower level of organic matter. Organic matter is a crucial source of soil N for crop growth through gradual decay and mineralization in the soil (Mbindah, 2017).

Soil available phosphorous in the site was very low when compared to the ranges given by Hazelton and Murphy (2007). Low available phosphorous in the soil implies that the garlic crops which belongs to the onion family could experience poor root development, stunted growth and delay in crop maturity unless P is supplemented as either foliar spray or soil fertilizer (Mbindah, 2017). Inadequate soil P may inhibit cell division in the meristematic tissues. This may encourage premature cell differentiation within the root tip that results in inhibition of primary root growth of young flowering plants (Chacon *et al.*, 2011). Most vegetables benefit from phosphorous fertilization if the soil test is less than 35 – 40 ppm phosphorous using the Bray – Kurtz P_1 extraction method (Tadesse, 2015).

Soil exchangeable potassium of the study site was found to be low when compared to ratings given by Hazelton and Murphy (2007). According to Tadesse (2015) soils that are less than 85 ppm potassium, are categorized under low potassium content. Garlic prefers a fairly neutral pH ranging 6.0 – 7.0 and hence if the soil is too acidic or too alkaline it causes slowed growth and late maturity in garlic. Moreover, nitrogen decreases as soil acidity increases while it becomes available as soil alkalinity increases (Tadesse, 2015). Potassium is important for maintenance of turgor pressure, accumulation and transport of metabolic products in the plant in water stressed conditions (Bationo *et al.*, 2012). Thus, potassium is an essential nutrient for optimal garlic production and yields. Mageed *et al.* (2017) reported that application of higher levels of K fertilizer in calcareous soils in an arid area, significantly improved plant water status of soya beans. Sufficient soil exchangeable K may therefore significantly contribute in enabling garlic crop to cope with prolonged drought.

III. CONCLUSION

The chemical analysis of goat manure-based vermicompost results showed that it contained higher levels of macronutrients nitrogen, phosphorous and potassium. Hence, if prepared well and applied using the appropriate rates it can sustain higher garlic plant growth by providing the macronutrients nitrogen, phosphorous and potassium. The soil analysis report of the experimental sites before planting revealed that the soils of the study area have low inherent soil fertility in terms of nitrogen, phosphorous and potassium. Hence, there is need to supply organic manures like goat manure-based vermicompost to enhance soil fertility for improved garlic productivity.

ACKNOWLEDGEMENT

We could like to acknowledge Dr. Patrick T. Gicheru who is the centre Director (KALRO Embu) for allocating land where the experiment was set up.

REFERENCES

[1] Ahmadabadi, Z., Ghajar, S. M. and Rahimi, A. S. (2011). Effect of vermicompost on soil chemical and physical properties. *Science and Technology of Agriculture and Natural Resources, soil and water science, Fifteen year*(58), 125-137.

[2] Alsina, I., Dubova, L., Steinberga, V. and Gmizo, G. (2013). The effect of vermicompost applications on growth of radish. *Acta Horticulturae, 1013*, 359-365.

[3] Ansari, A. A. and Sukhraj, K. (2010). Effect of vermiwash and vermicompost on soil parameters and productivity of

okra (Abelmoschus esculentus) in Guyana. *African journal of Agricultural Research, 5*(14), 1794-1798.

[4] Arancon, N. Q., Edwards, C. A., Atiyeh, R. and Metzer, J. D. (2004). Effects of vermicomposts produced from food waste on the growth and yields of greenhouse peppers. *93*, 139–144.

[5] Atiyeh, R. M., Lee, S., Edwards, C. A., Arancon, N. Q. and Metzger J. D. (2002). The influence of humic acids derived from earthworms-processed organic wastes on plant growth. *Bioresource Technology, 84*, 7-14.

[6] Atiyeh, R. M., Subler, S., Edwards, C. V., Bachaman, G., Metzger, J. D. and Shuster, W. (2000). Effect of vermicompost and compost on plant growth in horticultural container media. *Pedobiologia, 44*, 579-590.

[7] Azarmi, R., Sharifi Ziveh, P. and Satari, M. R. (2008). Effect of vermicompost on growth, yield and nutrient status of tomato (Lycopersicum esculentum). *Pak. J. Biol. Sci, 1*(14), 1797-1802.

[8] Bationo, A., Fairhurst, T., Giller, K., Kelly, V., Lunduka, R., Mando, A., Mapfump, P., Oduor, G., Romney, D., Vanlauwe, B., Wairegi, L. and Zingore, S. (2012). Handbook for integrated soil fertility management. Africa Soil Health Consortium, (ASHC), Nairobi.

[9] Bremner, J. M. and Mulvaney C. S. (1982). *Nitrogen-total. In: Methods of Soil Analysis, part 2, Chemical and Microbiological Properties.* American Society of Agronomy.

[10] Chacon, L. A., Ibarra, L. E., Sanchez, C. L., Gutierrez A. D. and Herrera, E. L. (2011). Global expression pattern comparison between low phosphorous insensitive 4 and WT Arabidopsis reveals an important role of reactive oxygen species and jasmonic acid in the root tip response to phosphate starvation. *Plant signaling and behavior*, 6: 382-392.

[11] DAFF. (2017). *Department of Agriculture, Forestry and Fisheries*. Retrieved 2019, from Production guidelines for garlic: http://www.daff.gov.za/publications.

[12] Degwale, A. (2016). Effect of vermicompost on growth, yield and quality of garlic. *Journal of Natural Sciences Research, 6*(3), 51-63.

[13] Hazelton, P. and Murphy, B. (2007). *Interpreting soil test results.* Collingwood VIC 3066: CSIRO.

[14] Jackson, M. L. (1967). *Soil chemical analysis.* New Delhi: Prentice Hall of India.

[15] Jaetzold, R. and Schmidt, H. (1983). *Farm Management Handbook of Kenya* (Vol. II). Nairobi, Kenya: Ministry of Agriculture.

[16] Jones, J. B. (2001). *Laboratory guides for conducting soil tests and plant analysis.* Boca Raton: CRC press.

[17] Kisaka, O. M., Mucheru-Muna, M., Ngetich, F. K., Mugwe, J. N., Mugendi, D. and Mairura, F. (2015). Rainfall variability, drought characterization, and efficacy of rainfall data reconstruction: case of Eastern Kenya. *Advances in Meteorology, 2015*, 16 pages.

[18] Lalitha, R., Fathima, K. and Ismail, S. A. (2000). The impact of bio-pesticide and microbial fertilizers on productivity and growth of Abel-moschus esculentus. *Vasundhara: the Earth, 1-2*, 4-9.

[19] Lazcano, C. and Dominguez, J. (2011). The use of vermicompost in sustainable agriculture: Impact on plant growth and soil fertility. *Nova science publisher, Inc.* , ISBN 978-1-61324-785-3.

[20] Mageed, T. A., El-Sherif, A. M., Ali, M. M. and El-Wahed, M. H. (2017). Combined effect of deficit irrigation and potassium fertilizer on physiological response, plant water status and yield of soya bean in calcareous soil. *Archives of Agronomy and Soil science*, 63 (6): 827-840.

[21] Mbindah, B. (2017). Evaluation of the combined use of organic and inorganic fertilizers on bulb onion (*Allium cepa* L.) yields and forecast of potential onion yields under climate change scenarios in west ugenya sub-county, Kenya. *Research gate*, 1-84. Retrieved October 2019, from https://www.researchgate.net/publication/328811653

[22] Mbithi, M. A., Mwanarusi, S. and Mwangi, M. (2015). Effect of different rates of vermicompost on growth and yield of beetroot (Beta vulgaris L.). *Egerton J. Sci. & Technol., 15*(ISSN No. 2073 - 8277), 30-43.

[23] Mikile, Z. (2001). *The use and agronomic effectiveness of Kraal manures in the Transkei region of the Eastern Cape, South Africa. Unpublished MSc. Agric. Dissertation.* Alice, South Africa.: University of Fort Hare.

[24] Mohammad, M. F. and Moazzam, H. A. (2012). Onion yield, quality and storability as affected with different soil moisture and nitrogen regimes. *South Western Journal of Horticulture, Biology and Environment*, 3(2): 145 – 165.

[25] Olsen, S. R., Cole, C. V., Watanabe, F. S. and Dean, L. A. (1954). *Estimation of available phosphorus in soils by extraction with sodium bicarbonate.* USA: U.S. Dep. Agric. Circ.939, USA.

[26] Pascal, J., Plumere, T., Thu, T. D., Rumpel, C., Duc, T. T. and Orange, D. (2010). The rehabilitation of tropical soils using compost and vermicompost is affected by the presence of endogenic earthworms. *Appl Soil Ecol, 46*(1), 125–133.

[27] Ramnarain, Y. I., Ansari, A. A. and Ori, L. (2019). Vermicomposting of different organic materials using the epigeic earthworm Eisenia foetida. *International Journal of Recycling of Organic Waste in Agriculture, 8*(1), 23-36.

[28] Rekha, G. S., Kaleena, P. K., Elumalai, D., Srikumaran, M. P. and Maheswari, V. N. (2018). Effects of vermicompost and plant growth enhancers on the exo-morphological features of Capsicum annum (Linn.) Hepper. *International Journal of Recycling of Organic Waste in Agriculture, 7. 10.1007/s40093-017-0191-5.*

[29] Tadesse, A. A. (2015). *Growth and yield response of garlic (Allium sativum L.) varieties to nitrogen fertilizers rates at Gantaafeshum, Northern Ethiopia. MSc Thesis,* Haramaya University.

https://dx.doi.org/10.22161/ijeab.51.8 *ISSN: 2456-1878*

Analysis of the effect of credit on the crop output of rice farmers in Benue State, Nigeria

Okolo Samson Ayegba, Olotu Olafemi Ayopo

Department of Agribusiness; Federal University of Agriculture Makurdi
Corresponding author. Email: samdivine147@gmail.com

Abstract**—** *The study was conducted to analyze the effect of credit on crop output of rice farmers in Benue state, Nigeria. Random and stratified sampling method was used to select 236 respondents in the study area. Out of the selected respondents only 208 responded and submitted the administered well-structured questionnaires correctly. Therefore, the study was based on 208 primary data collected from registered Savings and Credit Cooperative (SACCO) members in the three Local Government Areas of Benue State. In this study the year 2011 was used for before credit was obtained and 2015 for after credit was obtained. Descriptive statistics Foster –Greer –Thorbecke (FGT) poverty measure and double difference estimator was used to analyze the data collected. The analysis showed that 60.2 % and 67.5% which is majority are male beneficiaries and non-beneficiaries of credit respectively. The active age of the rice farmers were 31-40years and 41-50years for beneficiaries and non-beneficiaries respectively. The result from poverty severity index showed that 3% of the beneficiaries constitute the poorest after obtaining credit and 5% of the non-beneficiaries constitute the poorest also in the same year 2015. The result of the double difference estimate showed that the SACCO credit had a positive effect on the crop output of the beneficiaries of the credit with crop output mean value of 1176.84kg. SACCO executives and the Government should develop strategies that will bring in more funding, loans and grants to the cooperative consequently enhance availability of credit to members. This will help members who are smallholder farmers to become big estate farm holders. It is also possible that more credit availability to members is a key to poverty reduction due to its positive effect on the crop output as seen in the study.*

Keywords**—Analysis; effect of credit; crop output; rice farmers; poverty status; Benue State.**

I. INTRODUCTION

Poverty is a major scourge on farmers and low level income earners in Benue state. Benue state is known to be a state engaging more than 70 percent of its population in agriculture; agriculture is the back bone of the economy of the state (*Ajaero, 2007). The performance in agriculture is relatively average and dwindling due to the poor agricultural finance.* The research on the poverty reduction among rice farmers is very important since rice is a major staple in the study area.

Rice is consumed by more than 4.8 billion people in 176 countries and is the most important food crop for over 2.89 billion people in Asia, over 40 million people in Africa and over 150.3 million people in America, (Biyi, 2005). According to Jones, (1995), rice is the second most important cereal in the world after wheat in terms of production; while Nigeria ranks the highest as both producer and consumer of rice in the West Africa sub region. Akande and Akpokodje (2003) opined that, since the mid-1970s, rice consumption in Nigeria has risen tremendously, at about 10% per annum due to changing preferences while domestic production has never been able to meet the demand leading to considerable imports which today stands at about 1,000,000 metric tons yearly. The imports are procured on the world market with Nigeria spending annually over US $300 million on rice imports alone. Similarly, Biyi (2005) observed that the annual domestic output of rice still hovers around 3 million metric tons, leaving the huge gap of about 2 million metric tons annually, a situation, which has continued to encourage dependence on importation. This calls for the need to finance the rice farmers via the umbrella of the savings and credit cooperatives. With adequate financing of the SACCO it is very possible to meet the demand for rice in Nigeria and subsequently reduce poverty from rice farm families.

Therefore, the need for farmers to come together and form an autonomous association of individuals, voluntarily united

https://dx.doi.org/10.22161/ijeab.51.8 *ISSN: 2456-1878*

to meet their common economic, social and cultural needs through a jointly-owned and democratically controlled enterprise (International cooperative alliance, 1996). According to the global Multidimensional Poverty Index, International Monetary Fund (IMF) report by the United Nations (2015) the national average of poverty rate is 46.0%, the national proportion of those living above the poverty line is 54%. Benue State ranked 24th amongst the states living above the poverty line with 40.8% above the line and about 59.2% living below the poverty line.

Savings and credit Cooperatives (SACCO) are important in the provision of financial and banking services to low income households who for economic reasons cannot be covered by the activities of formal banks and financial institutions (Mwakajuilo, 2011). SACCO performs three major functions in relations to its members and general economic development of the country. These functions are collecting savings, giving credit and giving financial and non- financial advice to its members in order to facilitate and ensure that SACCO members utilize the micro credit they have borrowed from SACCO.

In some cases, some government and private institutions may also give financial assistance to SACCO in order to enable them give micro credit to their members (Mwakajumilo, 2011). He further posited that the different activities done by households in both urban and rural areas also mean the existence of different SACCO with the aim of assisting the Government to reduce high level of poverty and income inequality in the society.

Unemployment breeds a lot of private and social consequences which are negative (Alam, Khalifa, and Shahjamal, 2009; Alam, 2009). These include poverty, crime, social inequality, loss of output, family disintegration, among others. Governments all over the world make concerted efforts to mitigate these problems (Alam, 2009). In Nigeria several efforts have been made to create jobs for the teaming able bodied people who are available for work but who are yet to find jobs (Goodluck, 2011). One key source of unemployment in Nigeria is dearth of capital required to combine with other factors of production, which are land, labor and entrepreneurship (Nieman, Hough, and Niewenhuizen, 2003). Although growth is critical for poverty reduction, focus on growth alone is not enough (Almas, 2013). Micro-lending has been considered as the latest panacea for poverty alleviation (Magbagbeola, Adetoso, and Owolabi, 2010). Cooperative societies all over the world have been seen as one of the ways of reaching out to the un-banked and the neglected in the society and not a few have come to see it as an alternative to the regular banking, since it, in most case provides members of the group with the financial incentives without the rigors usually experienced in banking halls (Adewakun, 2012). Traditional cooperatives are common throughout Nigeria, but these groups tend to be small, with a common bond based on membership of a kinship, societal and low professional group (Adewakun, 2012). Saving and credit cooperatives Societies are known to provide funding to their members at reasonable interest rate and without requirement of collateral. They are therefore vital organs for financing food crop production (Mavimbela, Masuku, and Belete, 2010).

Objectives of the study

i. Describe the socio economic characteristics of beneficiaries and non-beneficiaries of SACCO Credit in the study areas;
ii. Determine the poverty status of beneficiaries and non-beneficiaries of SACCO credit;
iii. Analyze the effect of credit on the crop output of beneficiaries and non-beneficiaries of SACCO credit;

Statement of Hypothesis

The use of credit had no significant effect on crop output of beneficiaries of SACCO credit.

II. METHODOLOGY

Study Area

Benue State, the State lies between Latitudes $6^0$25'N and $8^0$8'N of the equator and Longitudes $7^0$47' and 10°E. (Ministry of land and survey, 2016). It has a total land-area of about 33,955 square kilometers with a population of 4,253,641 (NPC, 2006), with an average population density of 99 persons per square kilometer. The State is blessed with a great loamy soil for agricultural activities. It is one of the 36 states of Nigeria, It comprises 23 Local Government Areas (LGAs) grouped into 3 agricultural zones; A, B, C, respectively. The major food crops produced are yam, rice, cassava, maize, soybean, sesame, cowpea and groundnut at subsistence level. At the end of 2011, the poverty rate of Benue State was estimated at 31.9% (National Bureau of Statistics, 2012). Meanwhile at the end of 2015 the poverty rate of Benue State was estimated to be 59.2% based on data collected between 2004 and 2014 (Multidimensional Poverty Index, 2015) published by the United Nations.

There are areas of low population density such as Guma, Gwer East, Ohimini, Katsina-ala, Apa, Logo, and Agatu, each with less than seventy persons per square kilometers, while Vandeikya, Okpokwu, Ogbadibo, Obi, and Gboko

have density ranging from 140 persons to 200 persons per square kilometer. Makurdi LGA has over 380 persons per square kilometers. The study used zones (A, B, C) in the State to ease sample design and research instrument distribution. Zone A had the following Local Government Areas: Katsina- ala, Konshisha, Kwande, Logo, Ukum, Ushongo,Vandeikya. Zone B comprises Buruku, Gboko, Guma, Gwer- West, Gwer and Makurdi LGAs. Lastly Zone C comprises Agatu, Apa, Obi, Oju, Ogbadibo, Okpokwu, Otukpo LGAs.

Population and Sampling Procedures

Three Local Government Areas where rice cultivation was considerably high were selected, each from an agricultural zone in the State. The questionnaire was distributed to few active rice cooperatives whose major focus was solely on rice farming. The active rice cooperatives are distributed in the three LGAs viz; 9 in Katsina Ala, 82 in Makurdi and 10 in Agatu respectively. From these we have the following population for each LGA who are active with members participation measured by their contributions; Katsina Ala- 423 cooperators, Makurdi- 621 cooperators and Agatu- 167 cooperators Desk officer rice cooperative societies BSMANR, (2017). From the cooperatives actively participating in rice farming, few cooperatives that were accessible filled the questionnaire distributed, the following sample frame were taken: Katsina Ala with 80 registered member rice farmers, Makurdi with 123 registered member rice farmers and Agatu with 63 registered member rice farmers all with beneficiaries and non-beneficiaries inclusive respectively. A random sampling technique was used to select respondents for this study. The first stage was done by the selection of these three (3) local government areas because of the availability of more members of Savings and Credit Cooperative (SACCO) with documented records among the three agricultural zones of the state. At the end of the questionnaire administration, out of the 236 questionnaire administered, 208 were correctly filled and returned. Therefore the analysis was based on 208 completed rice farmers data collected. 128 of the beneficiaries and 80 of the non-beneficiaries of SACCO credit made up the 208 completed questionnaires.

Data Collection and Analysis

Primary data was used for this study. These were collected with the aid of structured questionnaire. Data was collected from 236 rice farmers using a structured questionnaire. Out of the 236 questionnaire 208 were retrieved correctly completed. Information collected include: the demographic details of beneficiaries and non-beneficiaries of SACCO credit. The outputs of the rice crop grown by the respondents were determined in Kilogram. Descriptive statistics to analyze the demography of the sample, Foster –Greer – Thorbecke (FGT) poverty measure was used to analyze the poverty status of respondents and the double difference estimator was used to analyze the effect of credit on farm output, t-test was used to test the hypothesis.

Model Specification

Double difference estimator

Information on both beneficiaries and non-beneficiaries was provided for before and after obtaining credit, it is literally a "difference of difference" (Albouy, 2010). The output of the rice crop grown by the respondents was determined in kg.

A positive mean double difference indicates a credit effect on beneficiaries, while a negative mean double difference indicates that the credit had no effect on beneficiaries (Nkonya *et al.*, 2008) the model is specified as:

$$DDE = \left[\left(\frac{1}{p}\sum_i^p(\bar{Y}_{tia} - \bar{Y}_{tib})\right) - \left(\frac{1}{c}\sum_j^c(\bar{Y}_{oja} - \bar{Y}_{ojb})\right)\right] \quad \text{(vii)}$$

Where;

$\bar{Y}_{tia} - \bar{Y}_{tib}$ = difference of mean crop output of beneficiaries after and before obtaining credit respectively.

$\bar{Y}_{oja} - \bar{Y}_{ojb}$ = difference of mean crop output of non - beneficiaries after and before obtaining credit respectively.

P= number of beneficiaries

C= number of non- beneficiaries

DDE = the difference between the mean changes in crop output for beneficiaries and non- beneficiaries. If the double difference estimates of the crop output of beneficiaries and non-beneficiaries of SACCO credit is a positive value. Then credit will have positive change on the rice output of beneficiaries.

III. RESULT AND DISCUSSION

Socio-Economic Characteristics of Respondents

An analysis of the sex of respondents indicated that majority (60.2 percent) where male, while 39.8 percent were females among the beneficiaries of SACCO credit. 67.5 percent and 32.5 percent were male and female non-beneficiaries of SACCO credit respectively. This could be because rice crop farming operations are so laborious such that the male who naturally are stronger seems to cope better than their female counterpart. The result agrees with the findings of Oguntola (1988) and Olorunsanya (2009) who concluded that farming

is male dominated profession and female are however more involved in processing of agricultural products. Another reason for the male dominance could be that most women in the study area do not take farming seriously like the males do. Therefore the SACCO is dominated by male since one of the major objectives of the participation in the rice farmers SACCO is to improve members' livelihood via upscale of rice production.

The result shows that most (48.4 percent) of the beneficiaries and (40 percent) of the non-beneficiaries were between the age of 31-40years for beneficiaries and 41-50years for non-beneficiaries respectively. The mean age of beneficiaries and non-beneficiaries is 45 years. With this age distribution, the indication is that majority of the beneficiaries and non-beneficiaries were within their active and productive working age. Therefore they participate and earn income from rice farming and other non-farming activities. This finding is in accordance with the findings of Windapo and Olewu (2001) and Bzugu *et al*., (2005) that productive and active persons participate more in agricultural and community development activities and groups such as SACCO.

Table 1 Socio-Economic Characteristics of the beneficiaries and Non-beneficiaries of credit (n=208)

	Beneficiaries		Non-beneficiaries	
Variables	**Frequencies**	**Percentages**	**Frequencies**	**Percentages**
Sex				
Male	77	60.2	54	67.5
Female	51	39.8	26	32.5
Total	**128**	**100**	**80**	**100**
Age (yrs)				
20-30	4	3.1	10	12.5
31-40	62	48.4	23	28.8
41-50	37	28.9	32	40.0
51-60	23	18.0	13	16.2
61-70	2	1.6	2	2.5
Total	**128**	**100**	**80**	**100**
Mean	45			
Marital Status				
Married	107	83.6	62	77.5
Single	21	16.4	18	22.5
Total	**128**	**100**	**80**	**100**
Education (yrs)				
0	10	7.8	10	12.5
1-6	43	33.6	31	38.8
7-12	60	46.9	27	33.8
13-18	15	11.7	12	15.0
Total	**128**	**100**	**80**	**100**
Hhsize				
1-5	44	34.4	16	20.0
6-10	73	57.0	49	61.2
11-15	8	6.2	6	7.5
16-20	3	2.3	9	11.2
Total	**128**	**100**	**80**	**100**

Source: computed from field survey data, 2017.

https://dx.doi.org/10.22161/ijeab.51.8 *ISSN: 2456-1878*

Poverty status of Beneficiaries and Non-beneficiaries of SACCO credit

The determination of poverty incidence index of beneficiaries and non-beneficiaries of SACCO credit a poverty threshold is established based on 2/3 and 1/3 mean per capita annual income (MPCFI) for beneficiaries and non-beneficiaries of SACCO credit. Further, 2011 was taken as the year before obtaining credit and 2015 as the year after obtaining credit.

The poverty depth index for the beneficiaries is 0.16 before obtaining credit and 0.07 after obtaining credit, while that of the non-beneficiaries is 0.17 in 2011and 0.11 in 2015. This implies that the non-beneficiaries had greater poverty depth index than the beneficiaries of SACCO credit. This can be said thus; the degree of poverty among non-beneficiaries was more compared to the beneficiaries. It therefore means that the farmers among the beneficiaries below the poverty line needs ₦20,347.6 annually which is 7 percent addition to their mean per capita annul farm income to attain the poverty line after obtaining credit. The non-beneficiaries will need 11 percent which can be translated to ₦31,974.8 annually addition to their mean per capita annual farm income to attain poverty line.

It was found that the beneficiaries had a poverty severity index of 0.13 and 0.03 before and after obtaining credit. While the non-beneficiaries had poverty severity index of 0.11 and 0.05 in 2011 and 2015 respectively. The result showed that the beneficiaries had a higher percentage of 13 percent of the poorest before obtaining credit in 2011and a lesser percentage of 3 percent after obtaining credit in 2015 indicating reduction in poverty severity. The non-beneficiaries had a poverty severity of showing that a higher percentage of 5 percent of poorest in 2015 compared to the beneficiaries. This implies that the credit obtained has a positive effect on the livelihood of the beneficiaries. It was found that poverty is marginally severe among respondents in the study area. About 3 percent of the beneficiaries constitute the poorest while about 5 percent of the non-beneficiaries constitute the poorest among the respondents. This is in tandem with the findings of Adebayo (2004) who reported that though the participating bee farmers had larger number of poor, the degree of poverty among the non-participating bee farmers was more when compared with the participating bee farmers and poverty is marginally severe among the non-participants.

Table 2: Poverty status of beneficiaries and non-beneficiaries of SACCO credit

Poverty Category	**Beneficiaries**		**Non-beneficiaries**	
	Before	After	Before	After
Non Poor	72	86	76	77
	(56.25)	(67.19)	(95)	(96.25)
Moderate poor	31	38	0	2
	(24.22)	(29.69)	(0.00)	(2.5)
Core Poor	25	4	4	1
	(19.53)	(3.13)	(5)	(1.25)
FGT Poverty Indices				
Poverty Incidence (Po)	0.44	0.33	0.05	0.04
Poverty Depth (P1)	0.16	0.07	0.17	0.11
Poverty Severity (P2)	0.13	0.03	0.11	0.05

Poverty Lines:	**Before**	**After**
MPCFI	**= ₦270,169.00 per annum**	**= ₦436,020.00 per**
2/3* (MPCFI)	**= ₦180, 112.00 per annum**	**= ₦290,680.00 per**
1/3* (MPCFI)	**= ₦90,056.00 per annum**	**= ₦145,340.00 per**

Source: computed from field survey 2017

https://dx.doi.org/10.22161/ijeab.51.8 *ISSN: 2456-1878*

Analysis of the Effect of Credit on Crop Output of Beneficiaries and Non-beneficiaries

Average output difference of the beneficiaries farmers was 3096.52kg and 6032.98kg before and after obtaining credit. Indicating that use of SACCO credit increased the crop output of beneficiary farmers. Furthermore, the average rice output for the beneficiary farmers was higher than that of non-beneficiaries after the SACCO intervention. The first single difference between after and before values is 2936.46kg. The mean output difference of the non-beneficiaries of the SACCO credit before credit is 3618.13kg and 5377.75kg after obtaining the credit. The first single difference between the crop output values of the non-beneficiaries before and after credit is 1759.62kg. The difference between the first differences of the beneficiaries and non-beneficiaries is the double difference value. This is the difference between the two outputs differences (2936.46-1759.52) is 1176.84kg. The result simply implies that the double difference analysis of the crop output of beneficiaries and non-beneficiaries of the SACCO credit is a positive figure. This result according to the a priori expectation indicated that the SACCO credit obtained by rice farmers in the study area had a positive effect on the crop output of the beneficiaries of the credit. Nkonya *et al*., (2008) posited that a positive double difference in output value indicates a positive effect of credit on beneficiaries output.

Table 3: Double difference estimates of the of credit on crop output of beneficiaries and non-beneficiaries of SACCO credit

Group	Crop Output		
	Before (kg)	After (kg)	Difference between Periods
Beneficiaries	3096.52	6032.98	2936.46
Non-beneficiaries	3618.13	5377.75	1759.62
Difference between groups	-521.61	655.23	1176.84

Source: field survey, 2017

The result of the hypothesis testing of crop output of the beneficiaries and non-beneficiaries showed that the F-value of the test had a positive value 21.197 and is statistically significant at one percent (1%). The mean of the crop output difference had a positive value 655.23, this indicate that there is a positive difference between the crop output of the beneficiaries and non-beneficiaries of SACCO credit. Therefore the null hypothesis which states that the use of credit has no significant effect on crop output of beneficiaries was rejected.

IV. CONCLUSION

The analysis revealed that Savings and Credit Cooperatives have helped the members improve their livelihood. It has positively changed members' poverty status by improving the crop output of beneficiaries of the SACCO credit. Nevertheless, the beneficiaries and non-beneficiaries had some factors that limited their efforts towards poverty reduction. There is room to the reduction of poverty via the improvement of the crop output of the SACCO members in the study area.

RECOMMENDATION

i. It is very important for families to encourage the female folks to participate in SACCO to improve their crop output and farm income. Some families with 8 member household sizes are still in core poverty due to inactive female folks in the household.
ii. SACCO executives and the Government should develop strategies that will bring in more funding; loans and grants to the cooperative consequently enhance availability of credit to members. This will help members who are smallholder farmers to become big estate farm holders. It is also vital to poverty reduction due to its positive effect on the crop output and increase in per capita annual farm income.
iii. Community members should be encouraged to get formal education and also adopt extension trainings for bumper crop output and to enable them benefit from the credit of the SACCO.

REFERENCES

[1] Adebayo, A. and Yusuf, O.R. (2004). Cooperatives and Poverty Alleviation in Rural Settlement of Kwara State, Nigeria.

[2] Adewakun, A. (2012), Cooperative as tool for enhancing financial inclusion, *African Newspaper of Nigeria*. Oct.13,P1.
[3] Ajaero C. (2007). A brand new image for Benue, *Newswatch Magazine* (Newswatch Communications). July 12,2007 from http://www.wikipedia.com
[4] Akende, S.O. and Akpokodje, G. (2003), Rice Prices and Market Integration in Selected Areas in Nigeria. *Agriculture and Rural Development Department Research Report*
[5] Albouy, David, 2010. Evaluating the efficiency and equity of federal fiscal equalization, Journal of Public Economics, Elsevier, vol. 96(9-10), pages 824-839.
[6] Alam G.M. (2009). Can governance and regulatory control ensure private Higher education as business or public goods in Bangladash? *African Journal Business Management*, 3(12), 890-906
[7] Almas H, 2013 The Relationship between Income Inequality and Globalization Retrieved July 7, 2010 from http://www.wider.unu.edu/
[8] Alam G.M., Khalifa. M.T.B and Shahajamal, M.M. (2009). Return from education system in Bangladash: an investigation on comparative flashback scenario. *African Journal Business Management*, 3(10): 567-575.
[9] Biyi, D. (2005). Government Policies and Competitiveness of Nigerian Rice Economy Paper represented at the *"workshop on Rice Policy and Food Security in sub-Saharan Africa"* organized by WADA, Cotonon, Republic of Benin, November 07 – 09, 2005.
[10] Bzugu P.M., M.M.Gwary, and Y.L. Idrisa, (2005).Impact of Extension Services on Rural Poverty Alleviation among Farmers in Askira/ Uba Local Government Area of Borno Sate. Shael Analyst, Faculty of Management Sciences, University of Maiduguri. PP96-103. Food Policy, 26, 4
[11] Goodluck, J. (2011). Job creation an urgent task; Punch Newspapers, Feb.24.
[12] Jones, M.P. (1995), the Rice Plant and its Environment West African Rice Development Association (WARDA) training Guide 2. P 1 – 16.
[13] Magbagbeola, J.A.O., Adetoso, J.A and Owolabi, O.A. (2010) Neglected and Apicrutilizes Species (NUS):A panacea for community focused development to poverty alleviation/poverty reduction in Nigeria. *Journal of Economics and international finance*, 2(10):208-211.
[14] Mavinbela, P., Masuku, M.B. and Belete, A. (2010). Contribution of savings and credit cooperatives to crop production in Swaziland: A case study of smallholder farmers. *African Journal of AgriculturalResearch;* 5(21):2868-2874.
[15] Mwakajumilo. S.L.I (2011). The Role of Informal Microfinance Institutions in Savings Mobilization, Investment and Poverty Reduction. A Case of Savings and Credit Cooperative Societies (SACCOS in Tanzania From 1961-2008. Unpublished Doctorial Thesis. St Clement University, Turks and Caicos Islands of British West.
[16] Niemen, G., Hough, J. and Niewenhunizen, C. (2003). *Entrepreneurship*- A South African Perspective. Hatfield, Pretoria: Van Schaik Publishers.
[17] Nkonya, E., Philip, D., Mogues, T., Pender, J., Yahaya, M. Adebowale, G.J and Arokoyo, T. (2008). Impact of a pro-poor community-driven development Project in Nigeria PP 10-36 *A report* submitted to International food policy research institute on sustainable solutions for ending hunger and poverty.
[18] Oguntola, S.I (1988). Female-Oriented Technologies in Agricultural Research in Oyo State. Unpublished B.Sc Thesis in the Department of Extension and Rural Development, University of Ibadan, Nigeria
[19] Olorunsanya, E.O (2009). Gender of Household Heads and Relative Poverty among Rural Farming Household in Kwara State. An Unpublished PhD Thesis in the Department of Agricultural Economics and Farm Management, University of Illorin, Nigeria

https://dx.doi.org/10.22161/ijeab.51.9 *ISSN: 2456-1878*

Comparison the Concentration of Purification Antigen MTSP11 and MPT63 as Serodiagnostic Active Tuberculosis

Ian Imanuel Fidhatami[1], Rosana Agus[2], Muh. Nasrum Massi[3]

[1]Graduate Student Department of Biology, Faculty of Mathematics and Natural Sciences, University of Hasanuddin, Makassar, Indonesia
[2]Lecturer in the Department of Biology, Faculty of Mathematics and Natural Sciences, University of Hasanuddin, Makassar, Indonesia
[3]Lecturer at the Faculty of Medicine, University of Hasanuddin, Makassar, Indonesia

***Abstract*—** *Tuberculosis remains a global concern in many countries because it is a contagious disease and become the second largest killer after HIV / AIDS. In suppressing the number of deaths from TB disease are increasing from year to year we conducted this test target antigen of Mycobacterium tuberculosis that is immunogenic by using molecular biology techniques.*

This study aims to the purification of recombinant proteins MTSP11 and MPT63 measuring the concentration of purified protein as a candidate serodiagnostic active tuberculosis. E. coli cells carrying the recombinant plasmid pQE30 Xa-Rv 3204 and pQE30 Xa-Rv 1926 cultured in LB medium containing ampicillin, then, do cell lysis. Further characterization of proteins and the last stage is the measurement results of protein purification.

The results obtained are MTSP11 protein has a molecular weight of 11 kDa while the MPT63 protein size of 16 kDa. The result of the calculation using the formula showed that the concentration of MTSP11 0.165423045mg / mL and the concentration of MPT63 is 0.155164115 mg / mL, the concentration of purified protein is best found in the last washing results by using the elution buffer.

Keywords*— *Mycobacterium tuberculosis, MTSP11, MPT63, SDS-PAGE.

I. INTRODUCTION

Tuberculosis (TB) is an infectious disease of the lung caused by *Mycobacterium tuberculosis* (Delogu, 2013). The slow progress in dealing with bacterial infections make the death rate is still relatively high (Liu, 2015).

World Health Organization, Defines the country with high load / high-burden countries (HBC) for TB based on three indicators of TB, TB / HIV and MDR-TB in which 48 countries entered into the list. One country may fall into one or both of the lists, even into the third. Indonesia along with 13 other countries, listed in HBC for all three indicators, meaning that Indonesia has a big problem in dealing with TB disease (WHO, 2017).

Pulmonary TB disease occurs when the immune system decreases. HIV-AIDS sufferers or people with poor nutritional status easier for infected and affected by TB. The percentage of TB patients who know their HIV status was among TB patients recorded an increase from 2009 by 2393 to 7796 in 2017 (Kemenkes RI, 2018). Tuberculosis children have special problems that are different from adults. TB children, the problems faced is the problem of diagnosis, treatment, prevention, and tuberculosis in HIV infection (Rahajoe, *et al*., 2005). During this time the diagnosis, treatment, prevention of tuberculosis in adults is more prioritized than in children, but the child is a higher risk group for immunity and is not fully developed (Walls and Shingadia, 2008).

The difficulty of diagnosing TB is still rising due to a variety of cases that are the resistant and the increasing burden of disease diagnostic tests indicates a need for inexpensive, reliable, easy to use and highly sensitive for TB cases. Utilization of specific *Mycobacterium tuberculosis* antigens and immunogenic as a candidate in the serodiagnostic test could lead to the proper diagnosis and rapid and immediate treatment.

In research Manca, *et al*., 1997 for MPT63 and Malaghini, *et al*., 2011 for MTSP11 discovered that the protein is an immunogenic protein from *Mycobacterium tuberculosis*, Two candidates protein produced by

https://dx.doi.org/10.22161/ijeab.51.9 *ISSN: 2456-1878*

Mycobacterium tuberculosis is important because it can interact directly with the host immune system so that the protein can activate the immune response in individuals infected with *Mycobacterium tuberculosis*.

Based on the above, to reduce the number of deaths from TB disease is increasing from year to year we conducted this test target antigens from *Mycobacterium tuberculosis*-reactive serum patients with active tuberculosis by performing the production of recombinant proteins, protein purification and measuring the absorbance value of the results of protein purification.

II. METHOD

Tool

The tools used are the incubator shaker (Heidolph Duomax 1030), centrifugation (Profuge), sonicator, Mini Protean II Bio-Rad, Elektroforator (Bio-rad), autoclave (Hirayama), laminar airflow (Esco Ductless Fume Cabinet), balance (Kern 440-47N), a micropipette (Bio-rad), flask (Pyrex), beaker (Pyrex), Eppendorf tubes (Axygen), Eppendorf tube rack (Biocision), falcon tube (BD), Freezer (GEA), measuring cup (Iwaki Pyrex), McCartney bottles, water bath (Memmert), and spectrophotometer cuvette.

Material

Materials used are clones of Escherichia coli BL21 which carries the recombinant plasmid pQE30 XA-Rv 3204 and pQE30 XA-Rv 1926 LB media (Luria Bertani), ampicillin, lysozyme, lysis buffer, wash buffer, buffer elution, Tris-HCl, Tris HCl, IPTG (isopropyl β-D-1-thiogalactopyranoside), Benzonase, Acrylamide/bis solution (Bio-rad), Sodium dodecyl sulfate (SDS), NaCl, KCl, Na2HPO4, KH2PO4, distilled, Loading buffer, Tris Base (Calbiochem), Ammonium persulfate, Tetramethylethylenediamine, Glisine, protein marker (Thermo Scientific), methanol (Merck), Glacial Acetic Acid, coomassie brilliant blue (Bio-rad), filter tip (Genfollower), Barrier tip (MultiGuard), Phosphate Buffered Saline (PBS) Marker proteins (*Precision Plus Protein ™ Dual Color Standards*), BSA (Bovine Serum Albumin).

2.1 Samples

Samples were taken from white colonies grown as a result of the transformation.

2.2 Culture of Recombinant Clones

Escherichia coli BL21 white colonies that carry the recombinant plasmid pQE30 Xa-Xa Rv pQE30-1926 and Rv 3204 by way were grown in LB medium (Luria Bertani).

2.3 Protein Production

Protein production was done by using sonication. The process by using ultrasonic waves in the frequency range 10 MHz KHz or known by the term ultrasonics.

2.4 Protein Purification

Protein purification using affinity chromatography column of Ni-NTA. Purification of recombinant proteins results then collected in Eppendorf tube to further analyzed by SDS-PAGE.

2.5 Protein Characterization

Characterization of protein by using technology most commonly used is the SDS-PAGE. Separation of high-resolution analysis of protein mixtures is sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE). Staining on electrophoresis results as much as 2x.

2.6 Protein Concentration Calculation

Calculation of protein concentration is calculated by using a spectrophotometer.

III. RESULTS AND DISCUSSION

3.1 Recombinant clones

The study began in October in Lab HUM-RC Wahidin Sudirohusodo Hospital. The clones that carry recombinant plasmids pQE30 Xa-Rv 3204 and pQE30 Xa-Rv 1926 comes from stock clones in an earlier study that examined in the laboratory HUM-RC RSWS. Reculture has done on farmed Luria liquid medium with the addition of *E. coli* bacteria that has brought the target gene and do well the addition of ampicillin. Transformant cells containing the recombinant plasmid will thrive on selection media containing ampicillin, while E. coli itself is sensitive to ampicillin. E. coli containing the recombinant plasmid can not grow on media selection (Mastutik, *et al*, 2015).

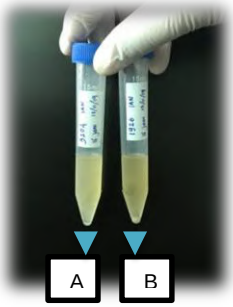


Fig.1: LB Media + Ampicillin + E. coli + IPTG
Description: A = Rv, 1926, B = Rv 3204

The culture media for 18 hours and then added with IPTG which serves to increase the production of recombinant proteins is desirable and will be marked with a thick band on a polyacrylamide gel. IPTG is a compound that has a

https://dx.doi.org/10.22161/ijeab.51.9 *ISSN: 2456-1878*

similar structure and function as an inducer lactose gene expression under the control of the promoter.

3.2 Protein Production

Bacterial culture in Eppendorf tubes in centrifuges to separate the supernatant (discarded) and the cell pellet. The addition of PBS buffer on the cell pellet serves as a solvent to assist in maintaining the consistency of the pH of the cells in maintaining the stability of proteins. Pellet cells that have been added to the physically broken PBS using sonication techniques in cold environments to keep the generated heat will not damage the protein that has been out of the bacterial cell.

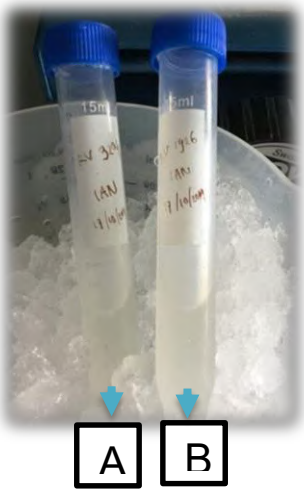


Fig.2. Results of sonication
Description: A = Rv, 1926, B = Rv 3204

3.3 Protein Purification

Sonication proteins purified using Ni-NTA Kit. The principle of purification of this kit is the addition of a buffer differently in the samples NPI-10 as a lysis buffer, NPI-20 as a wash buffer and NPI-500 as the elution buffer. Results obtained protein purification 3 stock code proteins by the addition of buffer. NPI-10 as a lysis buffer is the first washing in the purification of Ni-NTA affinity chromatography. Laundering of NPI-10 is called the Flow-through. NPI-20 as the wash buffer a second washing and the resulting sample in the form of protein, but not the targeted protein. Therefore do last washing with NPI-500 as the elution buffer. Affinity chromatography is used in natural immobilization of ligand by the target particles are then in to the columns. This column which will specifically bind to the desired target protein. In this case, the target protein is MTSP11 and MPT63.

3.4 Protein Characterization

Stock proteins were characterized using the SDS-PAGE method. The principle of the initial denaturation process involves the characterization of the protein components with an anionic detergent that also binds and then makes all the negatively charged proteins and proportional to the mass of the molecule. This step is followed by acrylamide gel electrophoresis through a porous matrix that separates proteins with a very good resolution based on molecular mass. Gel electrophoresis results later colored with a dye solution (staining) which aims to make the protein can bind the dye Coomassie Brilliant Blue. The second staining is done using the destaining solution to eliminate color on a gel that does not contain protein bands that were visible only on the gel that has a single protein thatwould establish a blue ribbon. Protein target is seen is the age of the gel with the code E.

Reading and determination of molecular weight protein purification results seen by marker proteins used. Results characterization weighs 11 kDa protein to protein MTSP11 and 16 kDa for the type of MPT63. The size of the target protein according to research conducted by (Lim, *et al*., 2004 and Siromolot *et al*., 2016).

3.5 Protein Concentration Calculation

The last stage of this study was the measurement of the concentration of each protein sample results from the characterization by using a spectrophotometer. The electromagnetic spectrum is divided into several areas of light. An area will be absorbed by atoms or molecules and the wavelength of light that is absorbed can show the structure of the compounds studied.

The electromagnetic spectrum covers a broad wavelength region of short wave-energy gamma rays high up in the micro wavelength. The Standard wavelength used is 500 nm. The main advantage spectrophotometric method is that this method provides a simple way to determine the quantity of the substance that is very small. Besides, the results are accurate, where the numbers recorded by the detector directly read and printed in the form of digital numbers or graphs that have regressed. The results of the calculation of the protein absorbance values presented in the table as below

Table 1. Results of Rv 3204 Spectrophotometer

Code	The absorbance value 500 nm	Result
Ft	0029	1,208
E1	0005	5,000
E2	0008	4,000
W1	0034	1,308
W2	0018	1,500

https://dx.doi.org/10.22161/ijeab.51.9 *ISSN: 2456-1878*

Table 2. Results of Rv 1926 Spectrophotometer

Code	The absorbance value 500 nm	Result
Ft	0008	1,600
E1	-0006	0750
E2	0013	1,000
W1	0022	1,571
W2	-0003	0429

Measurement of absorbance values that have been completed followed by a dilution of the BSA. BSA is a type of protein that has been used to compare the coating and protein concentration measurements of samples made. Dilution is carried out in stages from 10^{-1} to 10^{-5}.

Table 3. Dilution Spectro BSA

Dilution level	The absorbance value 500 nm
0	0156
0.1	0267
0.2	0:33
0.3	0:38
0.4	0405
0.5	0424

The measurement results in dilution of BSA is used to create a calibration curve equation and determine the best value for protein concentration

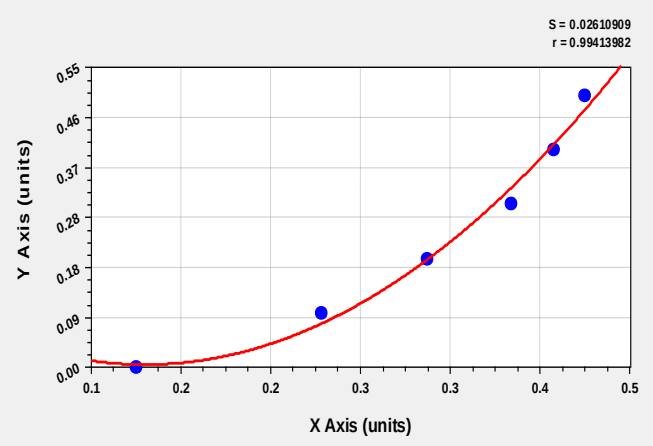


Fig.3: Calibration BSA

Absorbance value at MTSP11 and MPT63 protein compared to BSA absorbance values will result in the concentration of each recombinant protein and is calculated using the formula to the equation

y = a + bx + cx ^ 2 .... (1)

Description: a = 1.83E-01, -2.19E + b = 00, c = 6.80E + 00

Table 5. Results of Recombinant Protein Concentration Rv Rv 3204 and 1926

The sample code	Abs 500 nm	Concentration (mg / mL)
3204 Ft	0,029	0.124620324
W1	0,034	0.115789385
W2	0,018	0.145245182
E1	0,005	0.171741602
E2	0,008	0.165423045
1926 Ft	0,008	0.165423045
W1	0,022	0.137554835
W2	0,003	0.176021973
E1	0,006	0.169621816
E2	0,013	0.155164115

The result of the calculation using the formula shows that for MTSP11 protein, the highest concentration obtained is0.171741602 mg / mL and protein MPT63 with concentration values 0.169621816 mg / mL, high protein concentration which enables a high success rate also in serodiagnostic test.

IV. CONCLUSION

The conclusion of this analysis,

1. proteinMTSP11 has a molecular weight of 11 kDa while the MPT63 protein size of 16 kDa.
2. The highest concentration of the protein is MTSP110.171741602 mg / mL whereas with the MPT63 protein concentration values 0.169621816 mg / mL.

REFERENCES

[1] Delogu, G., Michela, S. and Giovanni, F. 2013. The Biology of Mycobacterium Tuberculosis Infection. Mediterranean Journal of Hematology and Infectious Diseases. (www.mjhid.org). ISSN 2035-3006.

[2] Kementerian Kesehatan Republik Indonesia. 2018. Data and Information Center (Indonesia Health Profile). The Ministry of Health of the Republic of Indonesia, Jakarta.

[3] Liu, W., Ying, XU, Hongbo, S., Jingran, Yan., Enzhuou, Y. and Honghai, W. 2017. Recombinant Bacille Calmette-Guérin coexpressing Ag85B-IFN-γ enhances the cell-mediated immunity in C57BL / 6 mice. Fudan University.

[4] Malaghini, M., Vanete, TS, Christian, MP, Marco, AK, Henrique, P., Afranio, K. and Carlos, RS 2011. Production for Recombinant Antigen Assays of Intradermoreaction for Diagnosis and Surveillance of Tuberculosis.*Journal of Biotechnology. 156: 56-58.*

[5] Manca, C., Constantine, L., Harald, GW, Donatella, U., Roberto, C. and Maria, LG 1997. Molecular Cloning, Purification and Serological characterization of MPT63, a Novel Antigen Secretd by Mycobacterium tuberculosis. American Society for Microbiology.*Journal of Infection and Immunity, 1997. Volume 2, 16 - 23. No. 1.*

[6] Mastutik, G., Reny, I'tishom, Sunaryo, Hardjowijoto and Suhartono, T. Son. Melanoma Antigen Gene Cloning 1 (MAGE-1) of the Testis Network to Get Plasmid Recombinant MAGE-1. Medical Faculty Airlangga University, Surabaya.

[7] Rahajoe, NN, Darfioes, B., Makmuri, MS and Cissy, BK 2005. National Guidelines for Tuberculosis Children. UKK Pulmonology IDAI PP, Jakarta.

[8] Siromolot, AA and Denis, VK 2018, a putative target antigens Cells of Mycobacterium tuberculosis MPT63 and MPT83. Pharmaceutical Research Journal of Biological and Chemical Sciences. Volume, 9. (2): 367-378.

[9] Walss, T. and Shigadia, D. 2007. The Epidemiology of Tuberculosis in Europe. Arch Dis Child.

[10] World Health Organization. 2017. Global Tuberculosis Report 2017 (www.who.int).

https://dx.doi.org/10.22161/ijeab.51.10 *ISSN: 2456-1878*

Selection of Lactic Acid Bacteria (LAB) Origin of Food Fermentation Probiotic Mixed as Candidate for Broiler

Aprisal[1], Yetti Marlida[2*], Husmaini[2], Harnentis[2]

[1]Magister Student at Graduate Program of Faculty of Animals Science at Universitas Andalas, Kampus Limau Manis, Padang, Indonesia, 25163

[2] Lecturer at Under Graduate of Faculty of Animal Science, Universitas Andalas, and Graduate Program at Universitas Andalas, Kampus Limau Manis, Padang, Indonesia, 25163

[2*]Corespondens Author : yettimarlida@yahoo.com / yettimarlida@ansci.unand.ac.id

Abstract— *This research aims to looking for 2 candidates from the new probioticlactic acid bacteria fermented food origin. This study uses seven isolates were isolated from whey, cassava, and fish budu. 7 isolates have been tested potential as a producer of glutamic acid but not known as a candidate probiotic petensinya. This study uses a completely randomized design, each repeated 3 times with 5 stages of the research are: 1) Testing capability isolates at low pH (pH 2.5); 2) testing the ability of isolates in bile (0.3 and 0.5%); 3) Viability isolates 4) Testing capability isolates in killing the pathogenic bacteria (Escherichia coli, Staphylococcus aureus and Salmonella enteritidis). The results showed the 7 isolates can be used as a probiotic candidates with the results of the resistance to pH 2.5 for 3 hours is 63.91-92.75% and the 6 hour incubation is 48,76-72.44%, resistance to bile salt concentration of 0.3% showed resistance to 22.86-83.57% and a concentration of 0.5% showed resistance 18.35-72,77%, 81.05-92.67% viability and inhibitory effect on the bacteria Escherichia coli is 7:33 to 12:01 mm, is 11:11 to 14:05 mm Staphylococcus aureus and Salmonella enteritidis is 12:02 to 18:08 mm. From 7 isolates after penseleksian ability as a candidate probiotic mixture then isolate F6 from durian fermented and isolate C8 from buffalo milk fermented (Dadih) are able to live at pH 2.5 for 3 hours of 92.75% and 86.06%, at 6 hours of 72.44 and 71.42%, the bile salt 0.3% were 83.57% and 78.75% and the concentration 0.5% are 77.17% and 72.77%, with viability were 92.67% and 92.23%, while the ability to kill pathogens such as Escherichia coli was 10.49 and 8.89 mm, Staphylococcus aureus was 14.05 and 13.70 mm, dan Salmonella enteritidis was 18.08 and 14.18 mm.*

Keywords— ***Lactic Acid Bacteria, Pathogenic Bacteria, Bile Salts, pH 2.5, Probiotics.***

I. INTRODUCTION

Antibiotic growth promoter is that antibiotics are often mixed in animal feed or drinking water. The use of antibiotic feed additives aimed at spurring growth, improving productivity, disease prevention, and feed efficiency. Usage of antibiotics to livestock has been ongoing since 1940 (Castañon 2007). The use of antibiotics in a sustainable and continuous endanger human health cause resistance among bacterial pathogens to antibiotics (Vignaroli et al., 2011) and the accumulation of residues of antibiotics in livestock meat (Tao et al., 2012). In Indonesia, the Ministry of Agriculture has issued Law No. Animal Husbandry and Animal Health 18 Year 2009 Article 22, Paragraph 4c which prohibits the use of antibiotics as feed additives (Ministry of Agriculture, 2009). The threat of a bad influence of antibiotics is also a serious concern to the researchers to conduct a search of alternative materials feed additives. One alternative feed additives that can be used are probiotic.

Probiotics are live microorganisms which when consumed can improve the health of livestock in a way to balance the microflora in the digestive tract when consumed in sufficient quantities. Probiotic bacteria are non-pathogenic microorganisms which, when consumed a positive influence on physiology and health of its host (Schrezenmeir and de Vrese, 2001). One type of bacteria that is often used as probiotics for livestock ungags are lactic acid bacteria (LAB). BAL is one of a group of bacteria that acts as a probiotic where these bacteria live in the digestive tract of

cattle. The use of lactic acid bacteria (LAB) as probiotic microorganisms has been ongoing since 1965 (Fuller, 1992).

Lactic acid bacteria (LAB) are a group of Gram positive cocci or rod-shaped, spore-forming, the optimum temperature ± 40 ° C, is generally not motile, anaerobic, catalase negative and positive oxidase, with lactic acid as the main product of carbohydrate fermentation.Lactic acid bacteria (LAB), which formed large potential used as a probiotic for lactic acid bacteria include microorganisms safe when added to food because it is not toxic and does not produce toxins, so-called food-grade microorganism known as microorganisms Generally Recognized As Safe (GRAS) is a microorganism which no risk to human health, even some types of bacteria are useful for human health (Kusmiati and Malik, 2002). In addition BAL also produces several compounds that function as antimicrobial mainly organic acids, hydrogen peroxide, and the fractions of proteins called bacteriocins (Ouwehand and Vesterland, 2004).

Lactic Acid Bacteria (LAB) has long been known for its role in the fermentation process that produces food products with different characteristics and flavor than fresh food. Some food fermentation produces lactic acid bacteria which are curd,cassava, and fish budu. Of the fermented food has been in isolation as much as 7 species of lactic acid bacteria. Seventh isolates have been tested using CaCO3 to determine that the ten isolates the lactic acid bacteria. The results of the test showed the ten isolates CaCO3 formed a clear zone disekitarannya, this means tersbut isolates classified as lactic acid bacteria (Maslami, 2019). Maslami (2019) have also proved that the seven isolates is glutamic acid-producing but not yet proven as a candidate probiotic.

FAO / WHO (2002) has set the minimum criteria that must be owned by probiotics as feed additives, namely 1) strain of probiotics should be identified characteristic phenotype or genotype, 2) In vitro assays, strains of probiotics should be able to live in the gastric acid and bile salts, able to stick to the mucus or intestinal epithelial cells, capable of producing antimicrobial that can suppress the growth of pathogenic bacteria, 3). Probiotic strains do not produce toxins, do not have properties resistant to antibiotics, and are not pathogenic.

The bacteria used as probiotics are still mostly given in the form of single or one type of strain. Still not much to look at giving some types of bacteria as probiotic mixture.The use of probiotic mixture to provide benefits more effective than the administration of one type of strain. This is in accordance with the opinion of Sanders and Veld (1999) which states that the use of multi-strain probiotics are more effective than single probitik, and more resistant to microbial infection.Effect of probiotic multistrain more effective as a spur growth of broiler (Timmerman et al., 2006).This was reinforced by previous researchers provide evidence that probiotics multistrain more effective than single strains of probiotics (Timmerman et al., 2004).Based on the above, then do research selection and characterization of fermented food BAL origin to get 2 isolates of LAB as a candidate probiotic mix for broilers.

II. MATERIALS AND METHODS

2.1. Material Research

Materials used in this study is 7 isolates of lactic acid bacteria (LAB) from fermented food. The ten isolates (B2, C8, C36, F6, I22, L15 and P1) has been electrically insulated and has been tested CaCO3 earlier, MRS Broth, HCl 37%, bile salts synthetic (oxgall), Nutrient Agar (NA), Escherichia coli , Staphylococcus aureus, Salmonella enteritidis, and others.

The tools used are bottle vial, a petri dish, vortex, test tubes, test tube rack, flask, beakers, micro pipette tip, *hockey stick*, Autoclaves, magnetic stirrer, hot-plate, Bunsen, laminar air flow, aluminum foil, Spectrophotometer, glass beaker, incubators, microscopes, pH meters, paper disk. Glass objects, needles ose, and others.

2.2. Research Implementation

This study was performed by using Experimental in Completely Randomized Designwith 7th isolates as treatment and each repeated 3 times.

2.2.1. Revitalization of Lactic Acid Bacteria

This research using 7 isolates were rejuvenated beforehand that aims to reactivate the stored isolates dilemari Cooling. Making the media for the rejuvenation of 7 isolates of lactic acid bacteria is by as much as 7.8 grams of MRS broth dissolved in 150 ml of distilled water. Then heated over a hot plate while stirring until homogenous. MRSB 10 ml of media was added to each vial bottle and then sterilized in the autoclave at a temperature of 121oC for 15 minutes. 1 ml lactic acid bacteria isolates taken using a micropipette and put into 10 ml MRS broth and incubated for 24 hours at a temperature of 37 ° C.

2.3. Observed variables

2.3.1. Selection of Lactic Acid Bacteria as a Probiotic Fermented Food Origin

Isolate Testing LAB (Lactic Acid Bacteria) made usingMRSB media (Man Ragosa Sharpe Broth) on various probiotic test (pH, bile salts and stickiness test). There are 7

https://dx.doi.org/10.22161/ijeab.51.10 *ISSN: 2456-1878*

isolates (B2, C8, C36, F6, I22, L15 and P1) were tested in various test Probiotics.

2.3.1.1. Gastric pH Resistance Test

The test method of resistance to gastric pH is based on the modified method of Hardiningsih et al. (2006). The test is performed using 10 isolates of lactic acid bacteria. The test is performed by using a medium MRS Broth 37% HCl is added to obtain a pH of 2.5 and a medium MRS Broth without the addition of HCl as a control with a pH of 6.8. Seterilisasi medium in the autoclave at a temperature of 121 ° C for 15 minutes. Bacterial isolates of 0.5 ml put in MRS Broth-HCl 5 ml and incubated for 3 and 6 hours at 37 ° C. Then read the absorbance at a wavelength of 600 nm. This study was conducted with three replications. Resilience isolates expressed as a percentage according Tokatli et al. (2015)

2.3.1.2. Bile Salt Resistance Test

The test method of resistance to bile salts was tested by the modified method of Vinderola and Reinheimer (2003). The test is performed using 10 isolates of lactic acid bacteria. Testing is done by adding a bile salt concentrations of 0%, 0.3%, and 0.5% in MRS broth medium. Medium sterilized by autoclave at a temperature of 121oC for 15 minutes. Bacterial isolates of 0.5 ml put in a 5 ml MRS broth that has been coupled with oxgall 0%, 0.3%, and 0.5% and incubated at 37 ° C for 5 hours. Controls containing MRS broth without addition of bile salt concentration (concentration 0%) compared with the treatment. Growth was measured by looking at the absorbance, at a wavelength of 600 nm. This study was conducted with three replications. Resilience isolates expressed as a percentage according Tokatli et al (2015).

2.3.1.3. Power Test Inhibitory Bacteria Against Pathogens

Test the antimicrobial activity of 10 isolates of lactic acid bacteria against pathogenic bacteria (Escherichia coli, Staphylococcus aureus and Salmonella enteritidis) is based on a modification of the method Awalia (2017), with the working procedures as follows:

1. To be prepared Nutrient media as much as 10 grams in 500 ml of distilled water, then homogenized and heated in a water bath, then in the autoclave.
2. After the media rather cold (± 45 ° C) added 0.2% of the bacteria that have been in the enrichment test 24 hours beforehand, homogenized, and then poured into a petri dish as much as ± 10 ml, cooled to solidify.
3. Meanwhile soak the paper disk in lactic acid bacteria for about 10 minutes.
4. After that solidifies paper disk placed on the surface of the medium NA which already contain pathogenic bacteria.
5. After that, incubated for 24 h at 37 ° C aerobically.
6. After 24 hours measured the diameter of inhibition zone is formed using a caliper.

According to Toy et al. (2015)

2.3.1.4. PowerTest Sticky or hydrophobicity

Hydrophobicity or attachment test was conducted using DarriDewanti and Wong (1995), using a stainless steel plate. Stainless steel is cleaned of dirt on its surface by soaking with a hot detergent solution (40-45OC) for 24 hours. Then the plates were rinsed with hot water (40-45OC) to not frothy and smooth, then dried. Once dry, the one marked stainless steel.

A total of 5.22 g MRS Broth was dissolved into 100 ml of distilled water as a medium for growth. Growth media and stainless steel autoclave at a temperature of 121oC for 15 minutes. Later stainless steel plate put in 25 ml MRS Brothyang isolates were inoculated with 1 ml of bacteria into erlemeyer, incubated for 24 hours at 37 ° C. Furthermore, by using a stainless steel surface swab wiped evenly. Swab inserted into tubes containing 10 ml phosphate buffer and homogenized. Then measured by the absorbance at a wavelength of 600 nm (A).

Measurement of growth in the liquid phase, in take 1 ml of media and diluted into 9 ml phosphate buffer solution. Then measured by the absorbance at a wavelength of 600 nm (Ao). The percentage of attachment can be calculated using the formula:

The diameter of inhibition zone = (Ao-A) X 100%
(Ao)

III. RESULTS AND DISCUSSION

3.1. Lactic Acid Bacteria Isolates resilience against Gastric pH

Based on probiotic on acid resistance test (pH) was performed using MRSB medium 0.1N HCl is added to obtain a pH of 2.5because the proventriculus and gizzard pH is 2.5-3.5(Surono, 2004).Results of lactic acid bacteria resistance to gastric pH can be seen in Table 1.

https://dx.doi.org/10.22161/ijeab.51.10 *ISSN: 2456-1878*

Table 1. The mean Resilience (%) of lactic acid bacteria to pH 2.5

Isolates	Incubation time	
	3 hours	6 hours
F6	92,75a	72,44a
C8	86,06b	71,42a
C36	73,52cd	48,94bc
B2	69,37de	52,90b
L15	67,49ef	50,98b
I21	75,66c	45,78c
P1	63,91f	48,76bc
SE	**1.625**	**1.307**

Note: superscript= different letters in the same column showhighly significant (P <0.01)
SE = Standard errer

Results of analysis of variance showed that each isolate LAB have different survival were significantly (P <0.01) on the condition of pH at 3 and 6 hours. F6 isolates have better survival against the acidic conditions at the time ingkubasi 3 hours and followed by C8 isolates with resistance level of 86.06%. The lowest resistance to acid conditions with a time of incubation for 3 hours ie P1 isolates (63.91%). Each LAB isolates decreased resistance at the time inkunasi 6 hours. Isolates F6 and C8 has the highest survival in acidic with 6-hour incubation period compared isolate C36, B2, L15, I21 and P1. Isolates with low resistance is I21 (45.78%). Table 1. Shows that viabililitas bacteria are incubated for 3 hours are from 63.91 to 92.75%.The results of this study are higher than research Badarinath et al. (2009) isolated from the okara has survival at pH 2.5 for 2 hours at 74.02%. While research Tokatli et al. (2015) lactic acid bacteria strain of Lactobacillus plantarum, Lactobacillus brevis and Pediococcus ethanolidurans isolated from traditional pickles own survival at pH 2.5 for 4 hours is 35-85%, 33-64% and 40-76%.

The results of the study Table 1 shows all the lactic acid bacteria can survive at pH 2.5 with a time of incubation for 3 hours and 6 hours, with minimal resistance ≥ 50%, which means that all isolates of lactic acid bacteria can be used as probiotics. As presented bySieladie et al. (2011)that the lactic acid bacteria as probiotics criteria can survive at low pH ≥ 50%. BAL able to survive at low pH conditions caused BAL has some physiological factors capable of regulating cell intracellular pH homeostasis(Van de Guchte et al. 2002; Cotter and Hill 2003).Fardiaz, (1989) describes that the Gram positive bacteria have a cell wall composed of 90% peptidoglycan and other thin layer is teikoat acid. Thick peptidoglycan and this teikoat acid chains that can maintain the shape of the cell wall of acidic extracellular conditions. BAL cell wall can retain its shape, so that the inside of the cell can still be protected.

Upon entering the digestive tract with a very acidic pH conditions, the bacteria can experience stress. Stress experienced is due to different intracellular pH with pH extracellular bacteria, so the lactic acid bacteria have to do in order to adapt the metabolism of intracellular to extracellular conditions.Siegumfeldt et al. (2000)explained that the lactic acid bacteria are able to maintain intracellular pH more alkaline than a pH of extracellular, but a decrease in intracellular pH persist with decreasing extracellular pH which supports tolerance to acid. Bacteria can degrade intracellular pH around neutral when the extracellular pH drops, but will use a lot of energy because of the difference of the proton gradient and result in the accumulation of organic acids anions that are toxic to the cell cytosol(Russell, 1992). Furthermore, also described by Siegumfeldt et al. (2000), lactic acid bacteria dynamic changes of intracellular pH due to a decrease in extracellular pH.

The food that goes into the stomach until it comes out of the stomach requires about 90 minutes of normal time, while the time used in this study was 180 and 360 minutes, or 4 times the minimum amount of time required to select BAL become probiotic bacteria. The aim is to ensure BAL really be able to survive in an acidic pH.

3.2. Lactic Acid Bacteria Isolates resilience Bile Salts

Lactic acid bacteria as probiotic candidate must be able to pass through the digestive tract trending conditions, in order to survive and grow. One is when bacteria enters the intestine where part or bile secreted in the gut.Journey probiotics in the digestive tract can face a variety of physiological conditions, therefore BAL as a candidate probiotic must be able to survive in various conditions of the upper digestive tract until reaching the intestine and can provide beneficial effects of probiotics on host (Babot et al. 2014). Resistance to bile salts is one of the most important selection criteria for probiotics because of the small intestine and the large intestine contains a high concentration of bile salts that are toxic to living cells (Floros et al., 2012). The concentration of bile salts in the duodenum and cecum chicken intestines are 0.008% and 0.175% (Lin et al., 2003).

Testing of lactic acid bacterial resistance to bile salts with a concentration of 0.3% and 0.5% for 5 hours, the result can be seen in Table 2.

Table 2. The mean percentage of lactic acidbacterial resistance to bile salts at concentrations of 0.3 and 0.5%

Isolates	Viability	
	0.3%	0.5%
F6	83,57a	77,17a
C8	78,75a	72,77a
C36	27,17cd	21,50b
B2	22,86d	18,35b
L15	24,65cd	23,57b
I21	31,39bc	21,96b
P1	32,34b	25,80b
SE	**2,175**	**2,884**

Note: superscript= different letters in the same column show highly significant (P <0.01)
SE = Standard errer

The results showed that each isolate LAB have different survival were significantly (P <0.01) against 0.3 and 0.5% concentration of bile salts. These results showed all isolates of lactic acid bacteria can withstand the bile salts with a resistance of> 20% and berpotesi as probiotics. In accordance with theBezkorovainy statement (2001) criteria for probiotics have an estimated survival rate of at least 20-40% of the strains selected. Isolates F6 da C8 is an isolate which has the highest resistance to the concentration of bile salts (oxgall) 0.35% ie 83.57 and 78.75%, namu peningktan already experienced a decrease in concentration of 0.5% bile salts (oxgall) to 77 , 17 and 72.77%. Increasing the concentration of bile salts can cause a decrease in the number of lactic acid bacterial colonies greater (Farida, 2006).The higher the concentration of bile salts, then the number of Lactobacillus cells that die will increase (Ngatirah et al., 2000; Kusumawati 2000).

The results obtained in this study compares favorably with the results obtained by Lee et al. (2014) that the survival rates of isolates of P. pentosaceus F66 by 26.6% with 0.3% bile salt with 2-hour incubation.Results of this study was higher than the results Melia (2018) isolates of lactic acid bacteria were isolated from buffalo milk can withstand the bile salts with a concentration of 0.3% for 5 hours by 40.58% and remained at 0.5% concentration of 35.22%.

Isolates F6 and C8 is also a difference isolates experienced a small decrease compared to isolates C36, B2, L15, I21 and P1.This small decline proves that isolates F6 and C8 have a great survival. As stated by Nurnaafi et al. (2015) which states that, in the great survival of lactic acid bacteria as probiotics are isolates which has a difference of a small decrease.

Lactic acid bacteria (LAB) were able to survive the conditions of bile salts caused by BAL enzyme bile salt hydrolases (BSH) which metabolize bile salts (Guo et al., 2010). BSH hydrolyze conjugated bile salts into conjugated bile salts that are not toxic for cells. Conjugated bile salts is bactericidal against sensitive microorganisms bile salts, can dissolve the lipid membranes and cause cell death (Begley et al. 2006). According to De Smet et al. (1995), Lactobacillus have an enzyme that can hydrolyze bile salts (bile salt hydrolase). This enzyme is able to modify the physical-chemical abilities possessed by bile salts that are not toxic to BAL.

Lactic acid bacteria also produce the enzyme β-galactosidase (Farida, 2006). Cells that die due to increased activity of the enzyme β-galactosidase to bile salts, the enzyme β-galactosidase, the lactic acid bacteria used to produce lactic acid from lactose (Surono, 2004). Increased activity of this enzyme is the bacterium attempts to adapt to the extracellular conditions, so that the speed of molecular diffusion of nutrients also increased and restrict the cell to control metabolism. Increased permeability of these cells results in intracellular material, such as cytoplasm and ribosomes extracted and undergo cell lysis. Though the cytoplasm serves as the venue for the hanpir all enzymatic reactions of cellular metabolism. In the cytoplasm, cell uses chemical energy to build and maintain cell structure and perform movements. Ribosomes synthesize proteins function. If this intracellular material, the bacteria can not metabolize, so it can not grow and even die (Lehninger, 2004).

3.3. Hidropobisitas power Lactic Acid Bacteria Isolates

Hydrophobicity of lactic acid bacteria can be seen in Table 3.

Table 3. Mean Hidropobisitas stickiness or lactic acid bacteria (%)

Isolate	Hidropobisitas
F6	92,67^{a}
C8	92,23^{a}
C36	89,72^{a}
B2	81,05^{c}
L15	89,29^{ab}
I21	86,42^{b}
P1	91,07^{a}
SE	**1,154**

Note: superscript= different letters in the same column show highly significant (P <0.01)
SE = Standard errer

https://dx.doi.org/10.22161/ijeab.51.10 *ISSN: 2456-1878*

Results of analysis of variance showed that each isolate different viability sanagat BAL had significantly (P <0.01). The average of the highest viability of 89.72 to 92.67%, ie isolates C36, P1, C8 and F6. These results indicate that each isolate has a high viability so that it can be used as a probiotic candidates. Because of the presence of BAL BAL attached to it is able to survive longer in the digestive tract that can further proliferate. Lactic acid bacteria are not able to stick with the good will come together intestinal peristalsis wasted leftovers for the next with feces. The next implication BAL were able to stick in the gut would be able to give the effect of intestinal defenses better to reject the possibility of pathogenic bacteria that are capable of adhering to the intestine.

3.4. Lactic Acid Bacteria Isolates Resilience Against Bacterial Pathogens

Criteria of lactic acid bacteria (LAB) used as a probiotic culture of which is its ability to inhibit pathogenic bacteria so that they can compete with pathogenic bacteria to maintain a balance of normal intestinal microflora (Salminen et al., 2004).

Diameter of clear zone formed as BAL antagonistic activity against pathogenic bacteria (Escherichia coli, Staphylococcus aureus and Salmonella enteritidis) can be seen in Table 4.

Table 4. The mean diameter of inhibition zone of lactic acid bacteria against pathogens

Isolate	Inhibition zone diameter (mm)		
	Escherichia coli	***Staphylococcus aureus***	***Salmonella enteritidis***
F6	10,49^{b}	14,05^{a}	18,08^{a}
C8	8,89^{c}	13,70^{ab}	14,18^{b}
C36	7,33^{d}	13,19^{ab}	12,10^{b}
B2	10,59^{b}	11,11^{d}	14,46^{b}
L15	10,92^{b}	11,28^{cd}	12,22^{b}
I21	12,01^{a}	12,31^{bcd}	12,02^{b}
P1	11,86^{a}	13,01^{bc}	12,87^{b}
SE	**.309**	**0.547**	**0.797**

Note: superscript= different letters in the same column show highly significant (P <0.01)

SE = Standard errer

The results of the study Table 4 shows the inhibition of lactic acid bacteria isolates against bacterial pathogens (Echerichia coli, Staphylococcus aureus and Salmonella enteritidis) highly significant (P <0.01). Isolates that have inhibitory Echerichia coli bacteria highest are I21 and P1 isolates with inhibition zone diameter of 12.01 and 11.86 mm. Average - Average Echerichia coli bacteria inhibition of each isolates in this study is 7.33 to 12.01 mm. These results are higher than the results obtained by Awalia (2017) the ability of lactic acid bacteria isolated from the gastrointestinal tract of chicken bangkok to inhibition*Escherichia coli*8.6 mm. The research Halim and Zubaidah (2013) the ability of lactic acid bacteria isolated from mustard sauce to the inhibition of Escherichia coli is 10.73 mm. Diameter inhibitory Lactobacillus plantarum to the bacterium Escherichia coli by 13.75 mm (Sunaryanto and Marwoto, 2012).

The diameter of inhibition zone of lactic acid bacteria isolates of Staphylococcus aureus against which the highest is 14.05 mm F6 isolates (Table 4). Visible diameter clear zone around the well, and showed antimicrobial activity generated by each of the lactic acid bacteria. Inhibition results in this study is higher than Awalia research (2017) with a diameter of 8.3 mm inhibition zone. The results ofresearch Halim and Zubaidah (2013), the ability to isolate lactic acid bacteria isolated from mustard sauce to the inhibition of Staphylococcus aureus was 10.68 mm.

Isoalat inhibition zone diameter of lactic acid bacteria against Salmonella enteritidis isolates highest are F6 ie 18:08 mm (Table 4). These results are higher than the results Chotiah and Damayanti (2018), in which the inhibition of bacteria Bifidobacterium Dentium against Salmonella enteritidis bacteria by 10 mm.

One mechanism in menghabat LAB isolates of pathogenic bacteria is an organic acid. Organic acids, in particular acetic acid and lactic acid has a strong inhibitory effect against Gram-negative bacteria and as a major antimicrobial compounds responsible for the inhibitory activity of probiotics against pathogens (Makras et al., 2006). Surono (2004) explains, the antimicrobial effect is the result of a decrease in pH and organic acids form undissociated. Undissociated acid can penetrate the cell wall of Gram-negative bacteria that have a high lipid content, ie 11-22%. Extracellular acidic conditions resulted in the pores of the cell wall of Gram-negative bacteria occur enlarged and cell permeability, so that many intracellular material out. Low extracellular pH conditions lead to acidification of the cell cytoplasm and acid that does not dissociate into lipophilic so that it can diffuse into the membrane. This situation would disrupt the transport system of the substrate in the cell, so that there is no supply of nutrients for cell metabolism processes.

Antimicrobial compounds produced by LAB in addition to the organic acid is a bacteriocins. The main target of bacteriocins produced is the cytoplasmic membrane, as bacteriocins initiate reactions that alter the permeability of the membrane that disrupts the transport of membrane or eliminate the kinetic energy of the protons that produce inhibition of energy production and biosynthesis of proteins or nucleic acids (Nissen-Meyer et al., 1992).

Gram negative bacteria are more sensitive to antimicrobial metabolites compared with Gram-positive bacteria. The lactic acid produced can weaken the permeability of Gram-negative bacteria by damaging the outer membrane of Gram-negative bacteria. Gram-negative bacteria do not have a proton pump mechanism which is able to balance the pH in cells and other antimicrobial substrate can not penetrate the cytoplasmic membrane (Cotter and Hill, 2003). The damage that occurs in the outer membrane of bacteria, occurs due to Gram-negative bacteria such as E. coli and salmonella has a thin layer of the cell wall and a thick lipid reaches 11-22%. Extracellular acidic state makes enlarged pores cells, increased cell permeability, so that the material is not required for entry into the cell metabolism, whereas many cell intracellular material coming out. Intracellular material such as cytoplasm and ribosomes can not perform its function to transport nutrients and synthesize proteins.

BAL resulting inhibition zone against bacterial pathogens third overall indicate that the antimicrobial compounds produced are able to inhibit the growth of pathogenic bacteria. Seventh lactic acid bacteria isolates had different inhibitory zone-beda terhadap bacterial pathogens, this is because each-masing isolates baktelactic acid ri mehave kCapacity of Different-beda in membunuh pathogenic bacteria.

IV. CONCLUSION

In concluded the ability as a candidate probiotic mixture then isolate F6 from durian fermented and isolate C8 from buffalo milk fermented (Dadih) are able to live at pH 2.5 for 3 hours of 92.75% and 86.06%, at 6 hours of 72.44 and 71.42%, the bile salt 0.3% were 83.57% and 78.75% and the concentration 0.5% are 77.17% and 72.77%, with viability were 92.67% and 92.23%, while the ability to kill pathogens such as Escherichia coli was 10.49 and 8.89 mm, Staphylococcus aureus was 14.05 and 13.70 mm, dan Salmonella enteritidis was 18.08 and 14.18 mm.

REFERENCES

[1] Awalia, F. 2017. Isolation and test the antibiotic activity of lactic acid bacteria in the intestine bangkok chicken gallus domesticus. Thesis. Faculty of Medicine and Health Sciences. Makasar.Makasar Alauddin State Islamic University.

[2] Babot J.D., E. Arganaz-Martinez, L.Saavedra, M.C. Apella, A.P.Chaia. 2014. Selection of indigenous lactic acid bacteria to reinforce the intestinal microbiota of newly hatched chicken - relevance of in vitro and ex vivo methods for strains characterization. Res Vet Sci. 97: 817.doi: 10.1016 / j.rvsc.2014.06.001.

[3] Begley, M., C.Hill, C.G.M.Gahan. 2006. Bile salt hydrolase activity in probiotics. Appl Environ Microbiol.73 (3): 1729-1738.doi: 10.1128 / AEM.72.3.17291738.

[4] Bezkorovainy, A. 2001. probiotics: determinants of survival and growth in the gut.Us National Library of Medicime National Institutes of Health. 73: 399-405.

[5] Cahyono, B. 2004.How to Improve Cultivation of Broiler.YayasanPustaka Nusantara, Yogyakarta.

[6] Castañon, J.I.R. 2007. History of the use of antibiotics as growth promoters in European poultry feeds. Poult Sci. 86: 2466-2471.

[7] Cotter, P.D., and C. Hill. 2003. Surviving the acid test: responses of gram-positive bacteria to low pH.MicrobiolMolBiol Rev 67 (3): 429453.doi: 10.1128 / MMBR.67.3.429-453.2003

[8] [FAO and WHO] Food and Agriculture Organization of the United Nations and the World Health Organization. 2002. Guidelines for evalution of probiotics in food. Report of the Joint FAO / WHO Working Group on drafting guidelines for the evaluation of probiotics in food. London Ontario (CN): FAO / WHO.

[9] [FAO / WHO] Food and Agriculture Organization / World Health Organization. 2006. probiotics in food: Health and nutritional properties and guidelines for evaluation. Rome: FAO / WHO.

[10] Fardiaz, S. 1989. Food Microbiology.Inter-University Center for Food and Nutrition.Bogor Agricultural Institute.

[11] Farida, E. 2006. Selection testing candidate probiotic lactic acid bacteria isolates the results of the local and its ability to inhibit the secretion of interleukin-8 from the flow cell 116. Thesis HCT.Postgraduate.Bogor Agricultural Institute.

[12] Floros, G., M. Hatzikamari, E. Lipoupou-Tzanetaki, and N. Tzanetakis. 2012. Probiotic and technological properties of facultatively heterofermentative Lactobacili from greek traditional cheeses. Food Biotechnology. 26 (1): 85-105.

[13] Fuller R. 1992.probiotics: The Scientific Basis. London (GB): Chapman & Hall.

[14] Fuller R. 1989.Probiotics in man and animals.Journal of Applied Bacteriology. 66, 365-378.

[15] Hardiningsih, R., and T. Yulinery Napitupulu RNR. 2006. Isolation and test the resistance of some isolates of lactobacilli at low pH. Biodiversity. 7 (1): 15-17.

[16] [Ministry of Agriculture] Ministry of Agriculture. 2009. Regulation of the Minister of Agriculture of the Republic of Indonesia Number 18 of 2009 on Livestock and Animal Health. Jakarta (ID): Ministry of Agriculture.

[17] Kusmiati and A. Malik. 2002. Activities bacteriocin from Leuconostoc mesenteroides bacteria pbac1 on various media. Makara, Health. 6 (1): 1-6.

[18] Kusumawati, N. 2002.Selection indigenus lactic acid bacteria as probiotics genus with the ability to maintain balance and reduce faecal microflora in rat blood serum cholesterol [thesis]. Bogor: Post Graduate Program, Bogor Agricultural University.

[19] Leeson, S. and JD Summer. 1996. Commercial Poultry Nutrition. 2nd Ed. University Books.University of Guelph. Guelph, Ontario, Canada.

[20] Lehninger, A.L. 2004.Fundamentals of Biochemistry.Volume 1. Translation: MaggyThenawidjaja. Erland, Jakarta.

[21] Lin, J., O. Sahin, L. O. Michel and Q. Zhang. 2003. Critical role of the multidrug efflux pump cmeabc in bile resistance and in vivo colonisation of campylobacter jejuni. Infection and Immunity. 71 (8): 4250-4259.

[22] Maslami, V. 2019. Isolation and seleksi origin lactic acid bacteriapwishful fermentation West Sumatra and a glutamic acid enghasilplikasinya in mningkatkan performans and carcass quality. disertasi. FacultyAgriculture. Andalas University.

[23] Melia, S. 2018. The potential of lactic acid bacteria isolates from buffalo milk as probiotics and bacteriocins producer for functional food. Dissertation. Faculty of Animal Husbandry. Andalas University.

[24] Ngatirah, E.,E.S.Harmayanti, and T. U. Rahayu. 2000. Selection of lactic acid bacteria as probiotic agents that have the potential to lower cholesterol. Proceedings of the National Seminar on Food Industry.

[25] Nurnaafi, A.I., Setyaningsih and Desniar. 2015. Potential probiotic lactic acid bacteria bekasam origin tilapia. Journal of Food Technology and Industry. 26 (1): 109-114.

[26] Russell, JB, 1992, Another Explanation for The Toxicitry of Fermentation Acid at Low pH: Anion Accumulation versus uncoupling, J. Appl. Bacterial 73: 363-370.

[27] Salminen, S., A.V. Wright, and A. Ouwehand. 2004. Lactic Acid Bacteria: Microbiology and Functional Aspects. 3rd edition. Revised and Expanded. New York: Marcel Dekker, Inc.

[28] Siegumfeldt, H., K.B. Rechinger, And M. Jakobsen. 2000. Dynamic changes of pH in individual intracelluler lactic acid bacterium cells in response to a rapid drop in extracellular pH. Appl. Environ. Microbiol. 66: 2330-2335

[29] Sieladie, D.V., N.F. Zambou, P.M. Kaktcham, A. and F. Cresci Fonteh. 2011. Probiotic properties of Lactobacillus strains isolated from raw cow milk in the western highlands of cameroon. Innovative Romanian Food Biotechnology. 9: 12-28.

[30] Simadibrata, M. 2010. Probiotics-Role in the World Medical.University of Indonesia. Jakarta.

[31] Surono, IS. 2004. Probiotics - Fermented Milk and Health. Jakarat (ID): Tri CiptaKarya.

[32] Tokatli, M., G. Gulgor, SB Elmaci, NA Isleyen and F. Ozcelik. 2015. In vitro properties of indegenenous of potential probiotic lactic acid bacteria. Biomed Reseach International: 1-8.

[33] Tao Y, Yu G, Chen D, Pan Y, Liu Z, Wei H, Peng D, Huang L, Wang Y, Yuan Z., 2012. Determination of 17 macrolide antibiotics and avermectins residues in meat with accelerated solvent extraction by liquid chromatography- tandem-mass spectrometry. J Chrom B. 897: 64-71. doi: 10.1016 / j.jchromb.2012.04.011.

[34] Timmerman, HM, CJ Koning, L. Murder, FM Rombouts, and AC Beymen. 2004. Monostrain, Multistrain and Multispecies probiotics: A Comparison of Functionality and Efficacy. Intr. Jour. Food Microbiol. 96: 219-233.

[35] Timmerman, HM, A. Veldman, E. van den Elsen, FM Rombouts and AC Beymen. 2006. Mortality and Growth Performance of Broiler Given Drinking Water supplemented with Chicken - Specific probiotics. J. Poult. Sci. 85: 1383-1388.

[36] Tokatli, M., G. Gulgor, SB Elmaci, NA Isleyen and F. Ozcelik. 2015. In vitro properties of indegenenous of potential probiotic lactic acid bacteria. Biomed Reseach International: 1-8.

[37] Van de Guchte M, Serror P, Chervaux C, Smokvina T, Emrlich SD, Marguin M. 2002. Stress responses in lactic acid bacteria. Antonie van Leeuwenhoek 82: 187-216.

[38] Vignaroli, C.,S. Pasquaroli, G. Zandri, L.F. Aquilanti. 2011. MultidrugresistantBiavasco enterococci in meat and animal faeces and co-transfer of resistance from an Enterococcus Durans to a human Enterococcus faecium.CurrMicrobiol. 62: 1438-1447.doi: 10.1007 / s00284-011-9880-x.

[39] Vinderola, C.G., And J.A. Reinheimer. 2003. Lactic acid starter and probiotic bacteria: a comperative "in vitro" study of the probiotic characteristics and biological barrier of resistance. Food Research International. 36: 859-904.

https://dx.doi.org/10.22161/ijeab.51.11 *ISSN: 2456-1878*

Survey of Apicultural Practices in Ibadan, Oyo State, Nigeria

A. A. Alarape[1*], M. K. A. Wahab[2], P. E. Arira[1]

[1*]Department of Wildlife and Ecotourism Management, University of Ibadan, Nigeria.
[2]Department of Wildlife and Ecotourism Management Osun State University, Osun State, Nigeria.
*alarapeabideen@gmail.com

Abstract*— *Apiculture is the practice and management of the bees in the hive with the sole aim of producing honey and other products including bee wax, propolis, bee-venom and royal jelly. Apiculture forms a line of productions that makes up agribusiness, but with challenges along the production chain. Little or no work has been done on the activities of beekeepers and the economic benefits of beekeeping in the study area. Therefore, the study aimed to assess the apicultural practices of beekeepers in Ibadan, Oyo State, Nigeria.

Structured questionnaires were administered to the beekeepers and honey marketers in three Local Government Areas (Akinyele, Ibadan North and Ibadan South East), purposively selected. The questionnaires comprise of two sections: demographics characteristics and apicultural practices of the respondents. The snowball sampling technique was used to select 30 beekeepers from the three Local Government Areas in the study areas. Data collected were analyzed using descriptive statistics such as mean, frequency table and percentage distribution.

The results of the study revealed that 96.7% were male dominated, within the age bracket (36.7%) and mostly married (76.7%). About 63.3% of the beekeepers use bee product as baiting material. At harvesting period, 80.0% of the respondents employ modern smoker for driving away bees while 56.7% stores honey in plastic as it provides suitable conditions for honey preservation. In line with their high education level, 86.7% of the respondents embrace modern beekeeping because of its being less tedious but with maximum output. Requiring low capital, modern beekeeping allows people an increased income resulting in improved standard of living. Many of the respondents, however, faced the problem of theft and bee sting,thus women were reluctant in practicing beekeeping as a means of livelihood. The paper recommended better security against thieves and more training on proper bee handling to minimize bee sting to encourage more people embrace the business with the hope of improved productivity.

Keywords*— *Apiculture, Baiting, Smoker, Bee-wax, Hive, Ibadan.

I. INTRODUCTION

Apiculture is the art and science of raising honeybees for man to benefit economically (Chinka, 1995). Beekeeping entails the practice and management of bees in hive (Ojeleye, 1999). The practice leads to the production of materials such as honey, bee-wax, bee-venom (Carauthersand Rodriguez, 1992). Beekeeping is a time tested (honoured) profession that not long ago provided almost the only source of concentrated sugar, although not only the sweetener for tropical people (Roubik, 1989). Beekeeping has evolved to be a very lucrative agricultural practice for local people in developing countries of the world (Serda*et al*., 2015). In Uganda, honey, bee-wax, propolis, royal jelly, bee venom is the major economic products(Herpburn and Radloff, 1998). Bees aid pollination of plants and servean ecological benefit (Delaplane, 2001). Originally, beekeeping is the keeping of bees in traditional hives built of any kind of suitable materials, that is locally available such as rock caves, cut timber etc. The more intensive beekeeping practice of the last century was based on the movable frame hives and virtually all the honey on the global market is supplied from this type of beekeeping (Krell, 1996).

Efficient utilization of honey bees requires information and knowledge of basic bee resources such as food plants and water. Hepbum and Radloff (1998) observe that detailed studies of honey bee-plant relationship are still patchy, making beekeeping less sustainable. Besides woody species as source of nectar and pollen in beekeeping, the role of herbs and shrubs is poorly understood and recognized by most beekeepers in Africa (Chinaka, 1995; Ojeleye, 1999).

https://dx.doi.org/10.22161/ijeab.51.11 *ISSN: 2456-1878*

Industrial zones or other areas with considerable air pollution causes high contamination of various hive products with dangerous or toxic chemicals (Mayer, 1997; Accorti, 1992). Agricultural use of poisonous chemical is another common source of contamination (Crane, 1990). The use of foul smelling chemicals to drive bees away at the time of harvest leave unpleasant flavour and odurs, and are absorbed by wax and honey e.g. nitrobenzene etc. (Daharu and Spoms, 1984). Alternatively, smoke can be used, but it has been established by Krell (1996) that excessive use of smoke during harvesting will flavour the honey quickly, no matter which smoker fuel was selected. Honey is extracted mostly by pressing, sometimes by dropping and by melting combs to separate wax from honey (Krell, 1996). The extracted honey can be purified by the removal of impurities such as wax particles and other debris incorporated during extraction. There are two practical methods of purification according to Krell (1996): settling and straining. The first entails leaving the honey in a large container, so that impurities can separate out based on the specific weight at temperatures of 25 to 30^0C. Straining can be used to supplement or complement settling especially in large processing plants. Strainers can be simple metallic screen, covered preferably with nylon mesh having holes of 0.1 to 0.2mm in diameter, at temperature close to 30^0C. Honey can be stored at a temperature of about 20^0C and a relative humidity of less than 65%. There is increasing loss in quality when stored at more than 25^0C, due to progressive chemical and enzymatic changes. Containers previously used for toxic chemicals, oils or petroleum products should not be used for keeping honey even after coating with paints as honey absorbs odours of all kinds which can be rapidly absorbed by a bee-wax coating and passed into the honey eventually (Krell, 1996). Honey production enriches human diet, improves income and revenue generation, provides job opportunities to many local people, and serves as raw materials for many industries, but its commercial production is hindered by problems in the study area. The rising demand for hive products and the rapid adoption of modern techniques of beekeeping notwithstanding, little or no work has been undertaken on the activities of beekeepers and the economic benefit of apiculture in the study area. It is necessary, therefore, to carry out a study on the activities of beekeepers in order to determine economic benefits of honey production, with a view to making recommendation for improvements. The results of the study will provide beekeepers with technical knowledge for optimal and more efficient operation. The result may also be useful to policy makers and agricultural workers, and also provide baseline information for further researches in the field. The paper therefore attempted to assess the apicultural practices of beekeepers in selected areas of Ibadan, Oyo State capital Nigeria.

II. METHODOLOGY

Study Area

Ibadan is the capital city of Oyo State, Southwestern part of Nigeria with a landmass of 3,080km^2 and a population of 2250000 people (NPC, 2006). It is located between latitude 7^0 23^1 47^{11}N and longitude 3^0 35^1 0^{11}E. The city is bordered by the Republic of Benin to the east, Lagos State to the north-east and Abuja to the south-west. Vegetation in Ibadan is typically a forest-savannah transition zone (Adekoya*et al.,* 2002). The mean annual rainfall is about 1420mm, while the temperature is relatively constant between 21^0C in the wet season and 27^0C during the dry season. During harmattan, a dry, cold and dusty wind blows between November and February. People of different ethnic groups reside in Ibadan with the Yoruba as the dominant inhabitants.

Data Collection/Sampling Techniques

Structured questionnaires were used to collect primary data from the beekeepers. This made possible collection of information on beekeeping and its economic benefit including the method of beekeeping used, harvesting and storing of honey. The questionnaires were administered by direct contact with the beekeepers. This made data collection to be more thorough as the majority of the respondents are barely literate.

The snowball sampling technique was adopted for the research to select one hundred respondents from the three Local Government Areas (Akinyele, Ibadan North and Ibadan South East). This choice was informed because beekeepers are difficult to access individually in the study area.

Data Analysis

The data collected were subjected to descriptive statistical analysis which included frequency and percentage distribution.

https://dx.doi.org/10.22161/ijeab.51.11 *ISSN: 2456-1878*

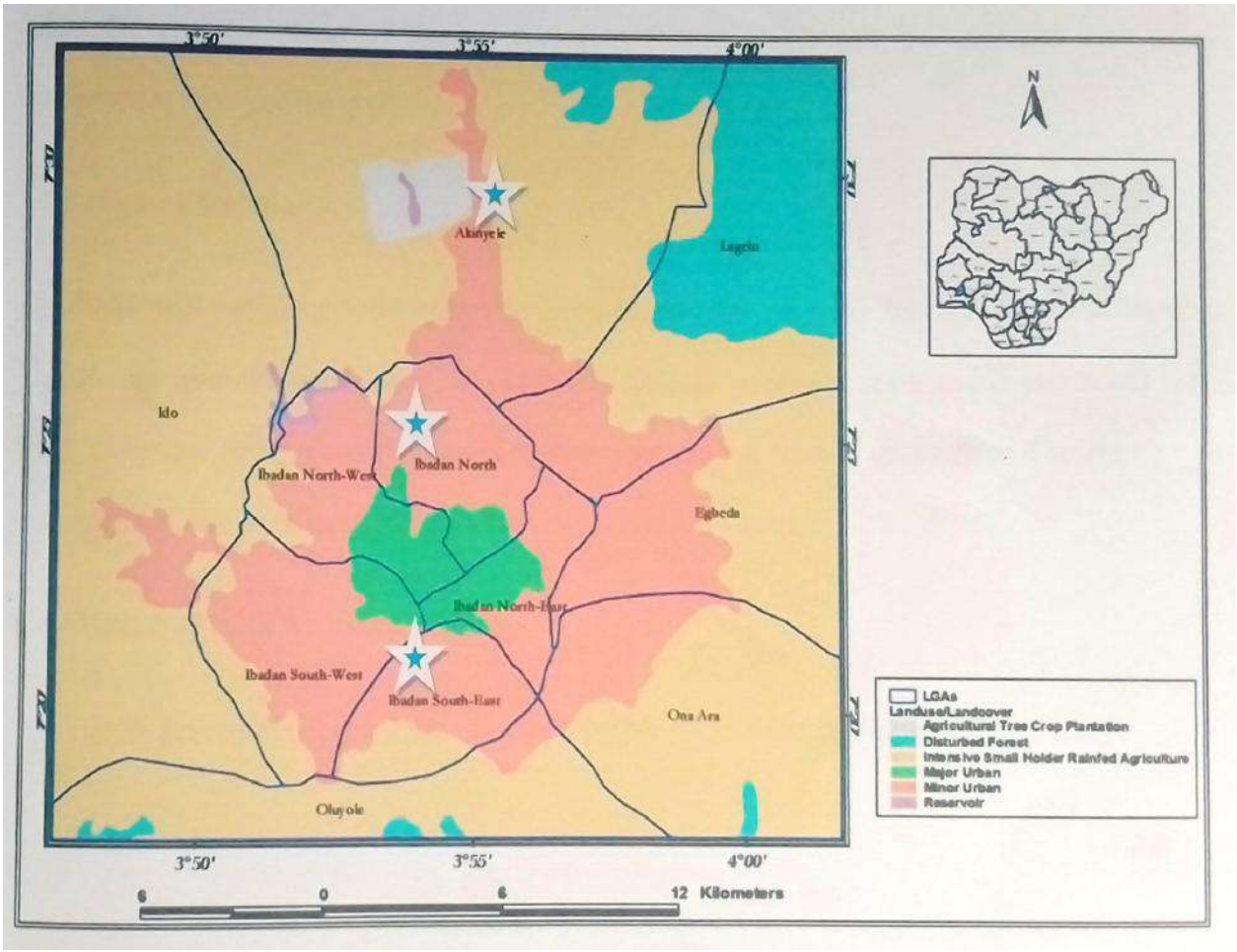


Fig.1: Map of Oyo State showing the Local Governmment Areas of Ibadan where the study was conducted. Sources: Adopted from speakersoffice.gov.ng, 2012

III. RESULTS AND DISCUSSION

Socio-economic characteristics of beekeepers

The socio-economic characteristics of the respondents considered are: sex, age distribution, marital status, household size, educational background and primary occupation. Table 1revealed that male (96.7%) were dominate and the highest percentage (36.7%) of the respondents fall within the age bracket of 40-49 years as honey harvesting is demanding and needs strong and brave people. This corroborates the findings of Adenuga (1987) and Malik *et al*., 2016 who observed that age is one of the determining factors when it comes to developmental initiatives and activities. Also, 76.7% of all the beekeepers sampled are married male. This agrees with the tradition of the local people that mostly men are into physical agricultural production. Similarly, half of the respondents (50%) had secondary education. This accounts for why larger percentage (86.7%) of the practitioners adopted modern beekeeping techniques. Abdul-kadir (1997) who established that educated farmers enjoys greater advantage over those that are not, which makes them receptive to proven changes and new ideas, thus improving their productivity and income generation. The results also reveals that about 83.3% of the respondents have between 3 and 8 individuals as members of their household while 10% have more than 15 household members. This large household size is typical of rural setting whereby people rely on family labour to carry out their farming activities. It was also shown that 60% of the surveyed beekeepers are artisan, alongside honey harvesting. This is because beekeeping serves as alternative income besides other enterprises.

Table 1: Socio-economic information of the beekeepers

Demographics	Frequency	Percentage %
Sex		
Male	29	96.7
Female	1	3.3
Age group (yrs)		
20-29	4	13.3
30-39	5	16.7
40-49	11	36.7
50-59	7	23.3
Above 60	3	10.0
Marital status		
Married	23	76.7
Single	6	20.0
Divorced	1	3.3
Level of education		
Primary	4	13.3
Secondary	15	50.0
Qu'ranic	3	10.0
Tertiary	8	26.7
Household size		
3-5	12	40.0
6-8	13	43.3
10-12	2	6.7
Above 15	3	10.0
Religion		
Christianity	21	70.0
Islam	9	30.0
Primary occupation		
Artisan	18	60.0
Professional	12	40.0

Methods of Beekeeping

The methods of beekeeping are classified in two including modern and traditional methods. Table 2 reveals that 86.7% of the beekeepers employed the modern method of beekeeping. This can be attributed to their being fairly educated which helped them recognize the superiority and advantages of using modern beekeeping equipment compared to traditional methods in terms of quality and quantity of honey produced. This cooperates with the observation of Farinde*et al*., (2005), who stated that modern beekeeping leads to increase in income as its investment is low and does not need daily care.

Table 2: Methods of beekeeping

Methods	Frequency	Percentage (%)
Modern	26	86.7
Traditional	4	13.3

Baiting materials

The bating materials considered by beekeepers to attract bees to the hive include cow dung, fruit, bee product and perfume. The largest percentage (63.3%) of the respondents uses bee product to lure bees to the hives. This can be explained by the people's adoption of the modern beekeeping techniques which ensures minimal contamination of the hive products.

Table 3: Baiting materials used in beekeeping

Baiting material	Frequency	Percentage (%)
Cow dung	5	16.7
Fruit	4	13.3
Bee product	19	63.3
Perfume	2	6.7

Means of Smoking Bees

The means the smoking bees employed by the beekeepers at the time of honey harvesting are smoldering twigs/grasses and modern smoker. At the time of harvest, 80% of the respondents (Table 4) uses modern smoker for smoking bees to keep them quite. This is in support of Krell (1996) who observed that excessive use of smoke during harvesting will flavor the honey quickly, no matter which smoker fuel has been selected.

Table 4: Means of smoking bees

Means	Frequency	Percentage (%)
Smoldering twigs/grasses	6	2.0
Modern smoker	24	80.0

Storage Facilities

The facilities used for the storage of honey were examined and found to include plastic and bottle. About 56.7% of both the beekeepers and honey marketers stores honey in plastic containers, while 43.3% keeps theirs in bottle (Table 5). The use of plastic is preferred possibly because it has temperature and humid conditions that will not affect the comb quality.

Table 5: Honey storage facilities

Storage facility	Frequency	Percentage (%)
Plastic	17	56.7
Bottle	13	43.3

Years of experience and quantity (kg) of honey sold

Table 6 shows that 63.3% of the beekeepers surveyed have been practicing bee-honey harvesting for up to between 1 and 10 years while 6.7% have spent up to 40years. This may be

https://dx.doi.org/10.22161/ijeab.51.11 *ISSN: 2456-1878*

because the modern beekeeping techniques adopted has been less tedious but with more financial returns.

It was also revealed that about 6.6% of the respondents sell unit of their honey at prices lower than ₦1000 while 63.3% sells between 1500 and ₦2000 per unit of measurement. This indicates price per unit of honey is not fixed; beekeepers can sell at any price as determined by market forces. Thus, profits can be maximized from sales, provided the quality of the honey remains intact. Ajao and Oladimeji (2013) also reported the role of honey in livelihoods of rural dwellers.

Table 6: Years of experience and quality (kg) of honey sold

Years in business	**Frequency**	**Percentage (%)**
1-10	19	63.3
11-20	6	20
21-30	3	10
31-40	2	6.7
Selling price (₦/) unit		
Below 500	1	3.3
500-1000	1	3.3
1000-1500	6	30.0
1500-2000	19	63.3
Above 2000	3	10.0

IV. CONCLUSIONS AND RECOMMENDATIONS

Majority of the respondents engaged in beekeeping were male Muslims and Christians who were married. They used bee products as baiting materials and stored their honey in plastic containers. A larger proportion of them employed modern smokers for smoking bees during harvesting. The largest percentage of participants was reasonably educated and knowledgeable about modern beekeeping technologies, which they accepted massively. They recognized that modern beekeeping methods were less tedious and more productive. Major problems facing beekeepers in the study areas included sting from bees and thievery from marauders. Governments, especially at the state and local levels, should therefore intensify efforts in providing more training for the practitioners, standardizing hive products from commercial beekeeping and improving marketing channels for sales at competitive prices.

REFERENCES

[1] Abdul-Kadir, I. (1997). The impact of extension for forestry development with reference to Katsina State. Unpublished M.Sc. dissertation. Department of Agricultural Economic and Extension. Usmanu Danfodiyo University, Sokoto.

[2] Adekoya, A.E. Akonola, A. A. and Fashola, K. (2002). Beekeeping as an income generating activity: A case study of honey farmers in Ibadan metropolis, Oyo State. *African Journal of Livestock Extension* 1(2):21-27.

[3] Adenuga, B. O. (1994). Extension services for forestry envelopment with reference to Oyo State, Unpublished M.Sc dissertation in the Department of Agricultural Extension Services, University of Ibadan.

[4] Accorti, M. (1992).The influence of the environment on the behavior and biology of bees in environmental monitoring. In proc. of sympon, the honey bee as monitoring insect for agricultural pollution (1992), Florence. Pp41-53.

[5] Ajao, A. M. and Oladimeji, Y. U. (2013). Assessment of contribution of Apiculture practices to household income and poverity alleviation in Kwara State, Nigeria, *International Journal of science and technology* 4(4):687-697.

[6] Caruthers, I and Rodriguez, M. (1992).Tools for agriculture. Nottingham, United Kingdom intermediate technology publication. P.238.

[7] Chinka, C. (1995).Beekeeping Keeping Technology for Nigerian Farmers. Extension Bulletin. No. 3. ABU, Zaria. P.33.

[8] Daharu, P. A. and Spons, P. (1984).Phenol residue level in honey. J. Apic Res. 23(2): 110-113pp.

[9] Delaplane, K. S. (2001): Honey bees and beekeeping. Extension entomologists. College of Agriculture and Environmental Sciences. University of Gorgia.

[10] Farinde, A. J., Soyebo, K. O. and Oyedokun, M. O. (2005).Exploration of beekeeping as a coping strategy in a deregulated economy.*Journal of agricultural extension*, 8:76-83pp.

[11] Herpburn, H. R. and Radloff, S. E. (1998).Honeybee of Africa, Springer-Verlag, Berlin.Func eco. 10:81-88.

[12] Krell, R. (1996). Value added products from beekeeping. FAO. Apicultural services bulletin No. 124. Rome.

[13] Malik, R. Yager, G. O. and Ojo, V. I. (2016).Assessment of Illegal activities carried out by rural Dwellers in selected support Zone communities of Gashaka-Gumti National Park, Taraba State, Nigeria, Nigerian Journal of Agriculture, Food and Environment. 12(1):51-55.

[14] Meyer, P. (1977). Nuclear energy and environment. Kemtechnic. 19(1): pp. 9-13.

[15] National Population Census. 2006.

[16] Ojeleye, B. (1999). *Foundation of beekeeping in the tropics.* Ibadan. Centre for Bee Research and Development (CEBRAD) press. 225p.

[17] Roubik, D. W. (1989). *Ecology and Management of Tropical Bees.*New York Cambridge University Press. 205p.

[18] Serda, B. Zewudu, T. Dereje, M. and Aman, D. (2015). Beekeeping practices, Production Potential and Challenges of Beekeeping among Beekeepers in Haramaya District, Eastern Ethiopia, Journal of Vertinary Science and Technology 6(5):1-5.

https://dx.doi.org/10.22161/ijeab.51.12 ISSN: 2456-1878

Similarity Analysis of Robusta Coffee Plant (*Coffea robusta* L.) at Three Altitudes in Merangin District, Jambi Province

Acep Sopandi*, Zulfadly Sayrif, Reni Mayerni

Faculty of Agriculture, Andalas University, Padang
*Email: acep.umb@gmail.com

Abstract— *Coffee is one of the plantation crops that has been developed and has become a commodity that is taken into account in strengthening the country's foreign exchange. The purpose of this study is to identify the similarity of robusta coffee plants at three heights in Merangin Regency, Jambi Province. The study began in October-November 2019 in Merangin District, Jambi Province using the survey method. The value of the coefficient of similarity is at 33 % which indicates that the degree of similarity is small. Heavy wet per 100 pieces of ripe, long bean coffee, as well as weight per 100 grains of coffee robusta scale on levels of water 12%, shows increasingly higher altitude of the place is getting better.*

Keywords— ***Phenotype, commodity, diversity, similarity, morphology.***

I. INTRODUCTION

Coffee plant (*Coffea* sp.) Is one of the plantation crops that was developed since the Dutch colonialism. This plant has become a commodity that is taken into account in strengthening the country's foreign exchange. This can be seen from the data of production, exports, and the area of Indonesian coffee. In 2016 - 2017 Indonesian coffee production and export ranked 4th in the world under Brazil, Vietnam, and Colombia (ICO, 2017). The volume and value of Indonesia's coffee exports in the last 5 years (2013-2017) experienced fluctuations, where the highest achievement was in 2013 which was 534,023 tons with an export value of US $ 1,174,029,000, while the lowest export volume occurred in 2014 which was 384,618 tons with export value of US $ 1,039,341 (Directorate General of Plantation Ministry of Agriculture, 2018).

The development of the area of coffee plantations in Indonesia in the last 5 years (2014-2018) averaged 1,238,417 ha with an average production of 686,097 tons in the year -1 -. The amount is dominated by smallholder plantations at 96.18%, 1.84% owned by state plantations and 3.77% privately owned (Dirjend Perkebunan, 2018). As for production, it reaches 94.99% of smallholder plantations, 2.51% of State plantations, and 2.50% of private plantations (Directorate General of Plantations, Ministry of Agriculture, 2018).

The growth of world coffee consumption is faster than the growth of world coffee production in the period 2012-2017. Growth in world coffee production only reached 1.18 percent while world consumption growth doubled from its production. (GAEKI, 2014).

Coffee plants that develop in Indonesia consist of Arabica and Robusta coffee. Both of these coffees have a high level of demand compared to other types of coffee. However, both copies have several problems, especially in terms of productivity. (Directorate General of Plantations, Ministry of Agriculture, 2018).

In Jambi Province, the highest number of coffee areas and production was found in Merangin District, which was 41.40% and 50.13% of the total area and production of coffee plants in Jambi Province, while the second-largest was Kerinci District with a total of 28.88% and 29.85%, the rest was spread over 8 (eight) Regencies / Cities namely West Tanjung Jabung, East Tanjung Jabung, Tebo, Bungo, Batang Hari, Muaro Jambi, Sungai Penuh and Jambi City-. Of the two types of coffee developed in Jambi Province namely Robusta and Arabica, Robusta coffee dominates the area of coffee plants in Jambi Province at 94.24% of the total 26,660 ha (BPS Jambi, 2018).

In addition to productivity issues, other problems, especially plant pests (OPT), seed quality and coffee flavor are challenges for Indonesia. Arabica coffee is susceptible to leaf rust caused by the pathogen *Hemileia vastatrix*, especially at an altitude of 600 - 700 m above sea level. The vulnerability of Arabica coffee to leaf rust disease is a limiting factor for production because this coffee is only better planted at a height of more or equal to 1000 m above sea level. Robusta coffee has properties that are more

https://dx.doi.org/10.22161/ijeab.51.12 *ISSN: 2456-1878*

resistant to the pathogen *Hemileia vastatrix* , so that this coffee can be planted at an altitude of fewer than 1000 m above sea level and optimum at an altitude of 600 - 700 m above sea level. However, its flavor is not as good as Arabica coffee (Indrawanto, *et al* ., 2010). Also Besides, coffee borer (PBKo) is an important problem in coffee cultivation.

These problems can be overcome by searching for coffee germplasm that has the expected characteristics through a breeding program. Germplasm character evaluation is one of the important activities in the breeding program that identifies the character and kinship of germplasm with a different visual appearance so that it can facilitate genetic management. Morphological characterization also has a major role in efforts to conserve germplasm, so that the continuity of information on the diversity of coffee plants can be well established (Soeroso, 2012). These efforts can assist breeders in making wise selections to get the expected plants. The purpose of this study is to find out the morphological diversity and similarity of robusta coffee plants at three heights in Merangin Regency, Jambi Province.

II. RESEARCH METHODS

The study began in October-November 2019 in Merangin District, Jambi Province using the survey method. The materials used in this study include coffee plants and their parts such as stems, leaves, flowers, fruit, and seeds . While the tools used are the ruler, meter, digital calipers, plastic bags, envelopes, digital cameras, GPS (*Global Position System*), *Lux meters* , digital altimeters , *soil testers* , paper bells, ladders, knives, machetes, *Color Chart for Plant tissue* and stationeries (pens, pencils, sipidol, and paper) . Similarity analysis is performed using a statistical calculation program, the NTSYS Ver.2.02 program.

III. DISCUSSION RESULT

Analysis of similarity aims to determine the distance of flower similarity between the genotype of a plant by using the properties of morphological. The nature of morphologic use to the introduction and display the similarity in kind . Of the 54 accessions of crops of coffee robusta in three heights in the District Merangin that observed , analysis similarity with using program *Numerical Taxonomy and Multivariate Analysis System* version 2.10e. (NTSys) to produce robusta coffee grouping (Figure 1) . The analysis is used to determine distance relationship resemblance between the genotype of a plant using properties its morphology . The morphology can be used for recognition that illustrates similarities in types . The type that has a resemblance to close has many similarities between the type of the other (Balkaya *et al* . , 2009).

In dendogram 54 accessions of crops of coffee robusta in three heights in the Merangin District province of Jambi on the character of qualitative were compared to show the coefficient of similarity with the value of 33-100% (Figure 1). The similarity coefficient value is an indication of the level of similarity of a plant . The more small value of the coefficient of similarity indicates getting smaller also the level of similarity between genotype was compared . It is meant also the level of diversity among genotype was compared to getting high if the large securities similarity coefficient values indicate the greater the degree of similarity between plants being compared and the smaller the level of diversity . According to Cahyarini (2004), the value of the coefficient of similarity <60% means having similarity genetic that far and if the value of the coefficients similarity > 60% can be said to have a semblance of genetic were close . The according to Sukartini (2007), if an individual has a value of similarity that is small or relationship resemblance that far , then the people that have a variety of genes both for use activity breeding .

Differences in the appearance of the plant can be caused by the difference like the plant (genetic) or differences in the environment or to both mutual influences . To express its genetic in full , plants require state environmental optimum (Sitompul and Guritno , 1995).

On the value of the coefficient of 33% (Figure 1), accession is divided into two groups of primary , namely groups 1 and 2. Group 1 consisted of 42 accessions were divided into two subgroups , namely 1A consists of 40 (RR1, RR2, RR4, RR15, RR3, RR7, RR8, RR13, RM1, RM3, RM5, RM7, RM4, RM13, RM15, RM15, RM18, RM16, RR5, RR6, RR9, RR12, RR12, RR14, RT1, RT3, RT8, RT2, RT7, RT11, RT11 RT9, RT12, RT13, RT17, RM 6, RM9, RM10, RM11, RM17, and RM19) accessions and 1B consisting of 2 (RM20, RM21) accessions . In group 2 consisting of 12 accessions also divided into two subgroups namely 2A consisting of 10 (RR10, RR11, RM2, RM12, RM8, RT4, RT15, RT6, RT14, RT15) accessions and 2B consisting of 2 (RT16, RT18) accession (Table 3).

At the 100% coefficient value there are several grouping accessions, namely , RR1 with RR2, RR4, and RR15, R R3 with RR7, RR8, and RR13, RM1 with RM3, RM5 , and RM17, RM4 with RM14, RM13 with RM15 and RM18, RR5 with RR6, RR9, and RR12, RT1, with RT3, RT8, and RT10, RT2 with RT7, RT9 with RT12 and RT13, RM6 with RM9, RM10, RM11, RM17, and RM19,

https://dx.doi.org/10.22161/ijeab.51.12 *ISSN: 2456-1878*

RM20 with RM21, RM2 with RM12, RT4 with RT5 and RT6, RT14 with RT15 , RT16 with RT18. Accession are flocking to the value of the coefficient of 100% indicates there is a similarity of character shapes stipules , the shape of leaves , forms the tip of the leaf , form the base of the leaf , the color of the leaves of young , the surface of the leaf , shape the edges of leaves , the color of ripe fruit , the shape of fruit , and form bean coffee robusta at three height . *Clustering* is intended for classifying objects based on the similarity of characteristics in between the objects the object of the . Objects are in classification to the one more *clusters* (groups) so that objects that are in the cluster will have a semblance of one with the other (Santoso , 2014).

If the value of the coefficient of similarity approaching the numbers 1,0 (100%), the diversity of genetic getting lower which means that individuals are similar but slightly or even not occur variations in genes. Whereas if the similarity coefficient approaches 0.1 (10%), genetic diversity is high . Anas and Yoshida (2004) stated that the samples were located on a line that together tends to have a diversity of genetic were lower in samples such.

IV. FIGURES AND TABLES

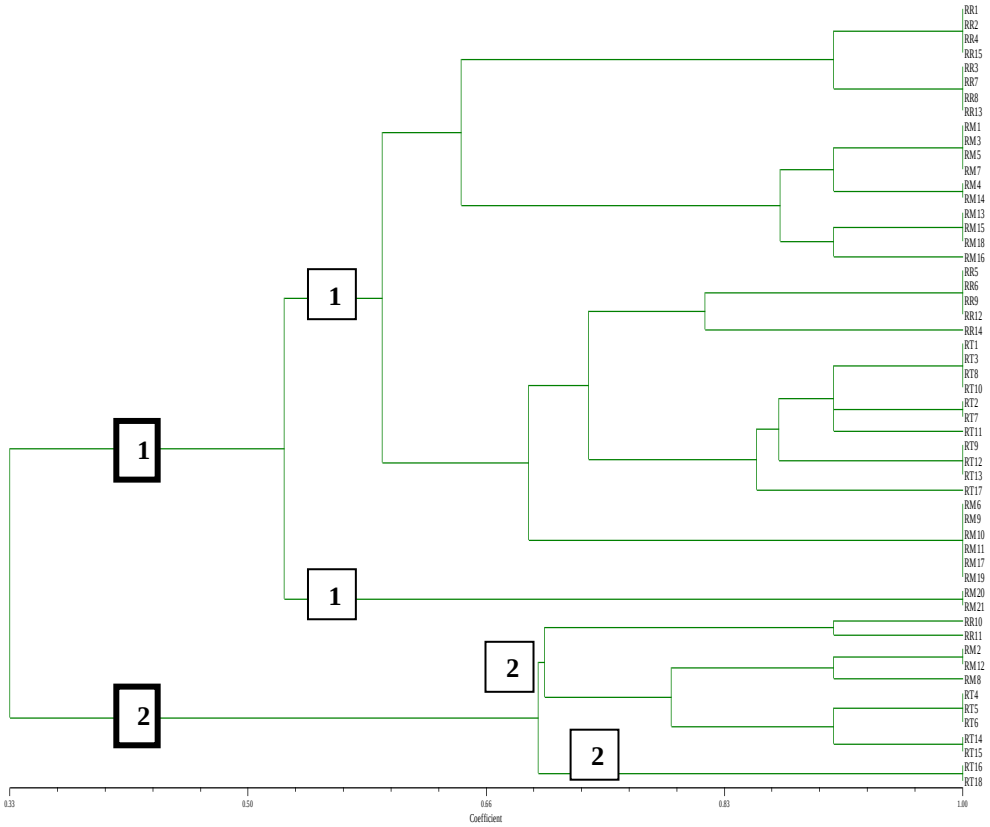


Fig.1: Dendogram 54 accessions of robusta coffee plants in three heights based on the character qualitative RR (Robusta plateau), RM (Robusta plateau Mediaum), RT (Robusta plateau)

Table.1: Grouping 54 Robusta Coffee Plants Accession at three altitudes in Merangin District Based on Qualitative Dendogram.

Main Group	Sub Group	Accession
1	1A	RR1, RR2, RR4, RR15, RR3, RR7, RR8, RR13, RM1, RM3, RM5, RM7, RM4, RM14, RM13, RM15, RM18, RM16, RR5, RR6, RR9, RR12, RR14, RT1, RT3, RT8, RT10, RT2, RT7, RT11, RT9, RT12, RT13, RT17, RM6, RM9, RM10, RM11, RM17, RM19
	1B	RM20, RM21
2	2A	RR10, RR11, RM2, RM12, RM8, RT4, RT5, RT6, RT14, RT15
	2B	RT16, RT18

V. CONCLUSION

The value of the coefficient of similarity is at 33 % which indicates that the degree of similarity is small. Wet weight per 100 ripe fruits ripe, long bean coffee, as well as weight per 100 grains of coffee robusta scale on levels of water 12% shows an increasingly higher altitude of the place is getting better.

REFERENCES

[1] Badan Pusat Statistik. 2018. Jambi Dalam Angka. Provinsi Jambi.

[2] Balkaya, A., R. Yanmaz, and M. Ozbakır. 2009. *Evaluation of variation in seed charactersin Turkish winter squash* (*Cucurbita maxima* Duch.) *populations*. N. Z. J.Crop Hortic. Sci. 37(3): 167-178.

[3] Direktorat Jenderal Perkebunan Kemeterian Pertanian Republik Indonesia. 2018. Statistik Perkebunan Kopi Indonesia 2017 – 2019. Jakarta

[4] [GAEKI] Gabungan Eksportir Kopi Indonesia. 2014. Perkembangan dan Tantangan Produk Kopi Olahan Indonesia www.gaeki.or.id. [Diakses 31 Agustus 2019].

[5] [ICO] *International Coffee Organization*. 2019. *Exports of all forms of coffee by exporting countries to all destinations* 2019. http://www.ico.org. [Diakses 31 Agustus 2019]

[6] Najiyati S, Danarti. 2012. Kopi, Budidaya dan Penanganan Lepas Panen. Jakarta(ID): PT. Penebar Swadaya.

[7] Soeroso SSDA. 2012. Pala (Myristica spp.) Maluku Utara berdasarkan keragaman morfologi, kandungan atsiri, pendugaan seks tanaman dan analisis marka SSR [disertasi]. Bogor(ID): Institut Pertanian Bogor

[8] Sitompul S.M, B. Guritno, 1995. Analisis Pertumbuhan Tanaman Talas. Gadjah Mada University Press. Yogyakarta. Hal 68-78

[9] Sudarsono, Ratnawati & Budiwati. (2005). Taksonomi Tumbuhan Tinggi. Universitas Negeri Malang. Malang.

[10] Sugiyono. 2007. Metode Penelitian Kuantitatif, Kualitatif dan R & D. Bandung: Alfabeta.

[11] Sukartini. 2007. Pengelompokkan Aksesi Pisang Menggunakan Karakter Morfologi IPGRI. J Hortikultura17(1):26-33

[12] Sumarno dan Zuraida, N. 2008. Pengeloaan Plasma Nutfah Tanaman Terintegrasi dengan Pemuliaan Tanaman. Pusat Penelitian dan Pengembangan dan Pangan Bogor. Buletin Plasma Nutfah Vol. XIV (2)

[13] Susilo AW. 2008. Ketahanan tanaman kopi (Cofffea Spp.) terhadap hama penggerek buah kopi (Hypothenemus hampei Ferr.). Review Penelitian Kopi dan Kakao. 24(1): 1-14.

[14] Stuessy, T.F. 1990. *Plant taxonomy*: *The systematic evaluation of comparative data*. Columbia University Press, New York: xvii + 514 hlm.

[15] Syukur M, S. Sujiprihati, dan R. Yunianti. 2012. Teknik Pemuliaan Tanaman. Penebar Swadaya. Jakarta.

[16] Tedianto. 2012. Karakteristik Labu Kuning (Cucurbita moschata) Berdasarkan Penanda Morfologi dan Kandungan Protein, Karbohidrat, Lemak pada Berbagai Ketinggian Tempat. (Tesis). Universitas Sebelas Maret. Surakarta.

[17] Van der Vossen, H.A.M., Soenaryo & Mawardi, S. (2000). Coffea L. dalam : van der Vossen, H.A.M. & Wessel, M. (Eds). *Plant Resources of South-East Asia* No.16: Stimulants. Backhuys Publisher, Leiden, The Netherlands. 6674.

[18] Van Steenis, C. G. G. J. Den Hoed. & Eyma, P. J. (2008). Flora Untuk Sekolah di Indonesia. Pradnya Paramita, Jakarta.

https://dx.doi.org/10.22161/ijeab.51.13 *ISSN: 2456-1878*

Diversity and Abundance of Corn Warehouse Pest Insect in Sumbawa District, West Nusa Tenggara

Muhammad Zulkarnain, Muhammad Sarjan*, Tarmizi

Masters in Dry Land Resources, Mataram University, Indonesia
* Coressponding Author: Muhammad Sarjan
Email: m_sarjan@unram.ac.id

Abstract*— *Warehouse pest insects can reduce the quality and quantity of corn stored in warehouses. This study aims to examine the diversity and abundance of warehouse pest insects that infest corn in the Sumbawa Regency warehouse. The study was conducted from May to September, located in Sumbawa Regency, covering the East, Central and West regions. The identification of pest insects was carried out at the Biotechnology Laboratory, Faculty of Agriculture and Biology Laboratory, Faculty of Mathematics and Natural Sciences, University of Mataram. The data is processed and reviewed using descriptive methods. The results showed that warehouse pest insects that infest corn in storage warehouses mostly came from the OrdoColoeptera and Hemiptera.

Keywords*— *Pest Insect, Corn Storehouse, Sumbawa Regency.

I. INTRODUCTION

Sumbawa Regency is one of the biggest corn producers in West Nusa Tenggara. Recorded in the data of the Agriculture Office of Sumbawa Regency (2018), Sumbawa Regency is able to produce corn nearly 60% higher than from 2012 to 2016, or with an average production achievement of 299,048 tons in that year.

High corn production in Sumbawa Regency requires storage warehouses during the post-harvest period. Warehouse as a storage place for corn commodity is one of the post-harvest supporting technologies before food arrives in the hands of consumers. The risk that must be faced while corn is in storage during storage is a decrease in quality and quality caused by insect pests. The decline in the quality and quality of corn can cause no small losses (Halid and Yudawinata, 1983). According to Morallo&Rejesus (1978) in Wahyuningsih (2000), damage to food stored in warehouses can result in losses of up to 5% to 10%. This statement was reinforced by Christensen and Kauffmann (1969) who suggested that of the estimated total loss of grain worldwide at least 50 percent was caused by insects.

Corn damage in storage is generally caused by infestation by pests. According to Rees (2004), there are a number of pest insects that infest storage material in a storage warehouse. The warehouse pest insects that infest generally come from the insects of the OrdoColeoptera and Lepidoptera, and the rest come from the OrdoOrthoptera and Psocoptera.

Quality assurance and quality of maize during storage at the warehouse needs to be done in Sumbawa Regency. One of the first steps to maintaining quality assurance is to obtain data on diversity and abundance of warehouse pest species through identification of pests in the storage warehouse. But to date, information and reports on pest insect species in storage warehouses in Sumbawa Regency have not been reported by all parties, either from academic researchers or the local Agriculture Service. Therefore, this research needs to be carried out to examine the diversity and abundance of warehouse pest insects that infest corn in the Sumbawa Regency corn storage warehouse.

II. MATERIALS AND METHOD

The study was conducted from May to September 2019 and was carried out in two stages, namely the field and the Laboratory. Field research was carried out for warehouse pest insect sampling which took place in three different regions. The three regions cover the East, Central and West Regions. There are two storage warehouses in each region, so we get six locations of the corn storage warehouse. The West Region consists of Rhee District with one warehouse (UD. Ai Salan) and one warehouse in Utan District (Abd. Gani's Warehouse), the Middle Region includes two warehouses in UnterIwes District (PT. Segar Agro Nusantara and CV. RestuSejati), and two corn warehouses in the Eastern Region including Plampang District (CV. FajarTerang and UD. Amanah). Data collection was carried out 4 times at 7-day

https://dx.doi.org/10.22161/ijeab.51.13 *ISSN: 2456-1878*

intervals to obtain data on diversity and abundance of corn barn pest insects. Data collection at the study site was carried out in 3 ways, namely sampling of corn infested with pest insects on the staple and warehouse floor, as well as making insect trap traps (Light Trap, Yellow Sticky Trap, Bait Trap and Pitfall Trap).

Laboratory research is carried out to identify insect pest sheds. The identification process was carried out at the Biotechnology Laboratory, Faculty of Agriculture and Biology Laboratory, Faculty of Mathematics and Natural Sciences, University of Mataram. Corn barn pest insect identification is carried out based on morphological samples, namely based on color, body shape, antennae, size and shape of wings, and the number of tarsals with the help of Borror identification books. The diversity and abundance of pest insect populations are calculated using the Shanon-Weiner formula with the equation H '= -∑ Pi ln Pi for insect diversity, and Pi = ni / N x 100% for insect abundance. The data obtained is then processed, reviewed and discussed descriptively.

III. RESULTS AND DISCUSSION

Diversity of Insect Pests in Corn Warehouses in Sumbawa Regency

The Ordo insects that were identified into the insect pests of corn warehouses in the three warehouse storage areas was found as many as 3 Ordo, including the OrdoColeoptera, Lepidoptera, and Psocoptera which were found in all storage warehouse areas. The OrdoColeoptera, which consists of 8 families, 11 genera and 12 species. Then followed by the Ordo Lepidoptera with 5 families, one genus and one species, and the OrdoPsocoptera with family, genus and species numbering one.

The same results were found in families, genera, and species for the Central and Eastern Regions, except the Western Region which showed different identification results from the two storage regions mentioned earlier. From a total of 12 families, 15 genera, and 16 species of insect maize pest identified in the Central and Eastern Region, not found 4 families from 12 families in the Western Region, namely Ptinidae, Silvanidae, Pyralidae, Psyllipsocidae, and one species (D. minutus) of the family Bostrichidae. In addition to these 4 families and one species, the same families, genera, and species found in the Central and Eastern regions (Table 2).

The West Region has fewer identified pests of corn warehouses compared to the other two regions (Table 2). The result was thought to be caused by the smallest size and size of the warehouse compared to the other two regions and the physical condition of the corn storage warehouse was quite good. The reverse results apply for the East and Central Region which has a warehouse with a larger size and relatively good physical condition.

The OrdoColeoptera is the most commonly found Ordo in all three warehouse areas (Table 2). This is because the insects of the Ordo are grain-eating insects. These results are consistent with the results of the research of Haines (1991) which mentions the ordoColeoptera is a postharvest insect pest that most damages grain storage materials such as corn compared to the Ordo Lepidoptera and Psicoptera. Sembel et al. (2002) corroborate previous reports that 10 species of pests were identified as grain-eating pests including Sithophilussp, Triboliumsp, Carpophilussp, Rhyzopertasp, Oryzaepilussp, Ahasverussp, and Criptoplestessp originating from the OrdoColeoptera. The OrdoColeoptera which attacks grain storage material is caused by the ability of insects from the ordo to adapt quickly to their environment. This is in accordance with the opinion of Wagiman (2015) which states that the OrdoColeoptera is a type of pest insect that is fast breeding and can adapt to the environment that is less supportive for its life cycle.

In total, there are 3 Ordo found in all three warehouse areas and 16 species are almost found in all three warehouse areas. The results show that species diversity is almost evenly distributed across the three regions. Based on the diversity index data shown in Table 1 explains that, the three regions have low species diversity because they have a diversity index value of less than 1 (H '<1). This condition reinforces that the diversity of insect pests in corn warehouses is almost equal in the three regions of the Sumbawa Regency warehouse. This happens because the environment is homogeneous in the three warehouse storage areas. Krebs (1978) explains that the more heterogeneous a physical environment the more complex the flora and fauna community in a place is scattered and the higher the diversity of its species.

Table 1. Corn Insect Diversity Index in Corn Storage Warehouses, Sumbawa Regency

Region	Diversity Index(H')
West	0.84
Middle	0.20
East	0.83

https://dx.doi.org/10.22161/ijeab.51.13 *ISSN: 2456-1878*

Tabel 2. Keragaman Serangga HamaGudang Jagung di Gudang PenyimpananKabupaten Sumbawa

Ordo	Family	Genus	Spesies	Found in Regional Storage Warehouse		
				West	Midle	East
Coleoptera	Bostrichidae	Rhyzopertha	*R. dominica*	+	+	+
		Dinoderus	*D. minutus*	-	+	+
	Curculionidae	Anthribidae	*A. fasciculatus*	+	+	+
		Sitophilus	*S. zeamais*	+	+	+
	Dermestidae	Attagenus	*A. fasciatus*	+	+	+
	Nitidulidae	Carpophilus	*C. lugubris*	+	+	+
			C. dimidiatus	+	+	+
	Ptinidae	Lasioderma	*L. serricorne*	-	+	+
	Silvanidae	Oryzaephilus	*O. surinamensis*	-	+	+
	Tenebrionidae	Tribolium	*T. castaneum*	+	+	+
		Alphitobius	*A. laevigatus*	+	+	+
Lepidopetra	Galeridae	Corcyra	*C. cephalonica*	+	+	+
	Pyralidae	Pyralis	*P. farinalis*	-	+	+
	Gelechiidae	Sitotroga	*S. cerealella*	+	+	+
	Oecophoridae	Endrosi	*E. sarcitrella*	+	+	+
Psocoptera	Psyllipsocidae	Liposcelidae	*Liposcelis*	-	+	+

The + and - signs indicate positive and negative pest insects found based on the corn storage area

An abundance of Insect Pests in the Corn Warehouse in Sumbawa Regency

1. Abundance of Pest Insects in the Western Region

There are two highest numbers of individual insect pests in the West, at the family and species levels obtained in the family Natidulidae, Carpophiluslugubris species with an abundance index of 72, 19%, and 19.12% in the Tenebrionidae family of the Triboliumcataneaum species. That is because both are from the same Ordo, the OrdoColeoptera which is known as a grain-eating insect. In addition to the Carpophiluslugubris and Triboliumcastaneum species of the families Natidulidae and Tenebrionidae, all other families and species have an abundance index of less than 10% (Table 3).

The two species above are pest species that are often found in storage sheds. Vega et al. (2019) states that Carpophiluslugubris is a primary pest in a storage warehouse originating from the OrdoColeoptera. While other reports mention that Triboliumcastaneum is called a pest insect that can damage storing materials up to 72% with a percentage decrease in the quantity of storing materials which ranges from 80%.

In theory, the increase in pest populations, especially warehouse pests, is influenced by several factors such as foodstuffs, storage microclimates, and the state of natural enemies (Yasin, 2009). Food material in the form of corn storage material that invites the two species into a storage warehouse. The temperature conditions in the storage shed also support the presence of the two species above, namely 26-30oC which is a range of temperatures that can still be tolerated by both species. Natural enemies are not found in storage areas in this region, whereas the presence of natural enemies can reduce the abundance of insect pest populations by carrying out their function as a biotic mortality factor so that the insect pest population can be kept at a low level (Luff, 1983).

Table 3. Post-harvest Corn Insect Abundance in the Western Region Corn Storage Warehouse, Sumbawa Regency

Ordo	Family	Species	Amount	Pi (%)
Coleoptera	Bostrichidae	*R. dominica*	72	0.68
	Curculionidae	*S. zeamais*	162	1.54
		A. fasciculatus	17	0.16
	Dermestidae	*A. fasciatus*	3	0.03

https://dx.doi.org/10.22161/ijeab.51.13 *ISSN: 2456-1878*

Ordo	Family	Species	Amount	Pi (%)
	Nitidulidae	*C. lugubris*	7674	72.98
		C. dimidiatus	530	5.04
	Tenebrionidae	*T. castaneum*	2010	19.12
		A. laevigatus	24	0.23
Lepidopetra	Galeridae	*C. cephalonica*	14	0.13
	Gelechiidae	*S. cerealella*	4	0.04
	Oecophoridae	*E. sarcitrella*	5	0.05

Pi: Pest Insect Abundance Index

2. Abundance of Pest Insects in the Central Region (Midle)

The highest population abundance index in the Central region was obtained in species of Liposcelis sp. from the Psyllipsocidae family, with an abundance index of 96.50%, while a population abundance index in species and other families has a very small abundance of less than 3%, as shown in Table 4.

The abundance of Liposcelis sp. in the Central Region it can be caused by the state of the micro environment that supports its growth and propagation. The humidity and temperature of the environment in the warehouse are at optimal conditions for the growth and development of Liposcelis sp., Ie the room temperature is approaching 30oC and the humidity nearing 60%. Leong (1995) reported that Liposcelis sp. can grow and develop optimally at a temperature of 30oC with a relative humidity of 75%. Liposcelis sp. is a warehouse pest which has the longest reported presence in a warehouse and causes a decrease in the quality and quantity of agricultural commodities (Turner, 1994).

Table 4. Post-harvest Corn Insect Abundance in Central Region Corn Storage Warehouse, Sumbawa Regency

Ordo	Family	Spesies	Jumlah	Pi (%)
Coleoptera	Bostrichidae	*R. dominica*	191	0.10
		D. minutus	5	0.00
	Curculionidae	*S. zeamais*	619	0.31
		A. fasciculatus	269	0.13
	Dermestidae	*A. fasciatus*	15	0.01
	Nitidulidae	*C. lugubris*	4308	2.16
		C. dimidiatus	406	0.20
	Ptinidae	*L. serricorne*	47	0.02
	Silvanidae	*O. surinamensis*	118	0.06
	Tenebrionidae	*T. castaneum*	728	0.36
		A. laevigatus	167	0.08
		C. cephalonica	53	0.03
Lepidopetra	Galeridae	*P. farinalis*	2	0.00
	Pyralidae	*S. cerealella*	36	0.02
	Gelechiidae	*E.sarcitrella*	20	0.01
Psocoptera	Psyllipsocidae	*Liposcelis*sp.	192669	96.50

Pi: Pest Insect Abundance Index

3. Abundance of Pest Insects in the Eastern Region

The highest population abundance index in the Eastern region comes from two species consisting of two different families (Table 5). The species is Liposcelis sp. from the Psyllipsocidae family, with an abundance index of 76.97%, and Carpophiluslugubris species from the Nitidulidae family with an abundance index of 11.74%. In addition to the two species of the two families, the population abundance index of the species and other families has a very small abundance, which is less than 10% as species of

https://dx.doi.org/10.22161/ijeab.51.13 *ISSN: 2456-1878*

Dinoderusminutus (Bostrichidae) and Endrosissarcirtella (Oecophoridae) with an abundance of 0, respectively, 02%, and Pyralisfarinalis (Pyralidae) with an abundance percentage of 0.00%.

Warehouse pest insects found in the Eastern Region are not much different from the other two regions. Liposcelis sp. previously found in the Central Region are also found in this region. The same applies to Carpophiluslugubris species that were previously found in the Western Region. Both species are found as the most abundant species in this region. This is presumably due to temperature and humidity factors which did not differ greatly in the three regions, although observations of minimum and maximum temperatures were found to be relatively different in the three regions. Temperature and humidity in the Eastern Region is very supportive for the proliferation of *Liposcelis sp*. which is in the temperature range of 27-31oC and relative humidity which ranges from 60-69%. The condition of the microenvironment is considered as the optimum condition for growth and development of the two dominant pest insect species found in this Region. Liposcelis sp. can grow and develop optimally at a temperature of 30oC with a relative humidity of 75% (Leong, 1995). Whereas Carpophiluslugubris is known as a warehouse pest that attacks grains such as maize commodities (Turner, 1994). In addition, the Carpophiluslugubris species is a primary pest in a storage warehouse originating from the OrdoColeoptera (Vega et al., 2019).

Table 5. Abundance of Corn Insect Pest in the Eastern Region, Sumbawa Regency

Ordo	Family	Species	Amount	Pi (%)
Coleoptera	Bostrichidae	*R. dominica*	354	1.18
		D. minutus	7	0.02
	Curculionidae	*S. zeamais*	319	1.07
		A. fasciculatus	139	0.46
	Dermestidae	*A. fasciatus*	35	0.12
	Nitidulidae	*C. lugubris*	3515	11.74
		C. dimidiatus	122	0.41
	Ptinidae	*L. serricorne*	6	0.02
	Silvanidae	*O. surinamensis*	35	0.12
	Tenebrionidae	*T. castaneum*	2256	7.54
		A. laevigatus	35	0.12
Lepidopetra	Galeridae	*C. cephalonica*	18	0.06
	Gelechiidae	*S. cerealella*	44	0.15
	Oecophoridae	*E. sarcitrella*	7	0.02
	Pyralidae	*P. farinalis*	1	0.00
Psocoptera	Psyllipsocidae	*Liposcelis*sp.	23040	76.97

Pi: Pest Insect Abundance Index

IV. CONCLUSION

The diversity of warehouse pest insects in Sumbawa is classified as low with a diversity index (H ') in the three regions of less than one (H' <1). The most commonly found Ordois the OrdoColeopteraand the least is Psocoptera. The abundance of corn barn pest insects is dominated by the OrdoPsocoptera (*Liposcelis sp.*) For the Central and Eastern regions with an abundance index of 96% and 76%, and the OrdoColeoptera (*Carpophiluslugubris*) for the Western region with an abundance index of 72%.

AKNOWLEGDEMENTS

This research was part of the first author Master Degree Project at Dryland Resource Management, University of Mataram. A million thanks to my lovely wife who always support me to do this research project.

REFRENCES

[1] Sumbawa Regency Agriculture Service. 2018. Data on Planting Area and Food Security Production. Sumbawa.

[2] Halid, H and Yudawinata. 1983. Types of BULOG Warehouse Storage Pests and Control Businesses. Paper

https://dx.doi.org/10.22161/ijeab.51.13 *ISSN: 2456-1878*

Presented at the Second Entomology Congress, January 24-26 1983, in Jakarta.

[3] Morallo&Rejesus. 1978. inWahyuningsih, S. 2000. Study of Insecticide Power of Pariah Seeds and Noni Seed Extract on the Development of Insect SitophiluszeamaisMotsch. Thesis. FATETA. Bogor Institute of Agriculture, Bogor.

[4] Christensen, C.M. and H.H. Kaufmann. 1969. Microflora. p. 158-191. In C.M. Cristensen (Ed.). Storage of Cereal Grains and Their Products. American Association of Cereal Chemist Inc., St. Paul.

[5] Rees, D. 2004. Insect of Stored Products.CSIRO Publishing. Australia. p.18.

[6] Haines, C. P. 1991. Insect and arachinids of tropical stored product their biology and identification. Natural resource institue, central avenue chatam maritime kentmey 4 TB. United Kingdom.

[7] Sembel, D.T, J. Rimbing, D. Kandowangko, 2002. Inventory and Identification of warehouse pests in grains as well as population development and loss and grain by pests. Collaborative Education for community Food Enterprice Development A&M University system and department of pests and plant diseases Faculty of Agriculture Unsrat.

[8] Wagiman, F.X. 2015. Postharvest Pest and Its Management. Second printing. GadjahMada University Press, Yogyakarta.

[9] Krebs, C.J. 1978. Ecological Methodology. New York: Harper and Row Publisher.

[10] Miriam Sanchez Vega, Alonso Mendez Lopez, Jose C. Salazar Torres, Aida I. Leal Robles, Sylvia Y. Martinez Amador, and Jose E. Perez Perez. 2019. Diversity of Insect Pest Damaging Quality of “Huitlacoche” (Corn Smut) at Saltillo, Coahuila, Mexico. Southwestern Entomologist. Vol 44, Issue 3, p.627-636.

[11] Yasin, M., 2009. Ability to Access Insect Powder Beetle Pests and Physiochemical Factors That Affect it. Proceedings of the 2009 national cereals seminar. Cereals research center. ISBN: 978-979-8940-27-9.

[12] Luff, M.L. 1983. The Potential of Predators for Pest Control. Agriculture, Ecosystem & Environment. Vol. 10, Issue 2, p.159-181.

[13] Turner, B. D. 1994. Liposcelis bostrichophila (Psocoptera: Liposcelididae), a stored food pest in the UK. International Journal of Pest Management, 40:2, p.179-190.

https://dx.doi.org/10.22161/ijeab.51.14 *ISSN: 2456-1878*

Assessment of Agricultural Credit Acquisition among Small Scale Poultry Farmers in Katsina-Ala and Konshisha Local Government Areas in Benue State, Nigeria

Ashikegh, S. A*[1]; Prof. Iheanacho, A. C[2]; Atser, E. A[3]

[1]Nigeria Security and Civil Defence Corps, Plateau State command, Jos.
E-mail:ashikeghstephen@gmail.com
[2] Department of Agribusiness, Federal University of Agriculture, Makurdi.
[3]Nigeria Agricultural Quarantine Service, Abuja.
*Correspondence Author

Abstract— *The objectives of this study were to assess agricultural credit acquisition among small scale poultry farmers in Katsina-Ala and Konshisha Local government areas in Benue State, Nigeria. The study adopted the survey research design, using the cross-sectional approach because it involves assessment of agricultural credit acquisition among small scale poultry farmers in the study area. Structured questionnaire was administered to eighty –eight (88) randomly selected respondents in the study area and interview technique was also used to obtained additional information from the respondents whenever the need arose. The study concludes that agricultural credit is adjudged as an important input to increase poultry production and poultry enterprise using loans in the study area is profitable; however the profitability level is a function of the scale of production. The study suggested that government should remove security advancement of collateral conditions that discourages poultry farmers from commercial banks facilities, formal and informal money lenders should reduce the interest rate to one digit for farmers to afford convenient repayment and more borrowers to be encouraged, and lenders should timely approve/disburse fund to farmers for effective acquisition and curtail the protocols involved in credit acquisition.*

Keywords— ***Credit, Acquisition, Poultry, Farmers.***

I. INTRODUCTION

Agriculture is the bedrock on which every successful and stable economy the world over is built. In Nigeria, agriculture accounts for one third of the Gross Domestic Product (GDP) and employs about two third of the labour force (Otunaiya *et al.,* 2012.). The challenges of food insecurity and hunger in developing countries like Nigeria have caught the attention of experts and governments worldwide (Emaikwu *et al*., 2011; FAO, 2003). Population growth, urbanization, and income improvements are the main drivers of increased demand for foods of animal origin in developing countries (Abdullah *et al*. 2011; Steinfeld, 2003). The sufficient supply of animal protein is most critical in the global food basket crisis (FAO, 1995). As a result, growing demand has led to a rise in the production of foods of animal origin all around the globe, especially from poultry and pigs (FAO, 2010).

What is known as poultry farming in Nigeria developed slowly but steadily from the 1950's until, when as a result of introduction of Structural Adjustment Programme (SAP), it began to experience the crisis which persisted until today (Okonjo, 2000). The importance of animal protein apart from its palatability is undisputed. Animal protein provides man with high quality food nutrients for growth and tissue replacement. It determines the level of nutrition of the population and the health of the work forces, which in turn determines the development of a nation and its economy.

The Nigerian government in an attempt to alleviate this problem has always resorted to mass importation of meat. The Obasanjo led administration has put more effort towards

https://dx.doi.org/10.22161/ijeab.51.14 *ISSN: 2456-1878*

self-sustainability in poultry production by placing ban on the importation of frozen chicken. The problem which solution is sought is that of harmonizing or balancing the shortage in supply caused by the ban on importation of frozen chicken. This can only be achieved by increasing poultry production. Also, many Nigerian livestock farmers in their own effort have preferred to invest in the poultry industry making it easy for the transformation of the previous back yard and less efficient system of poultry keeping to a more scientific system, despite their poor capital base. The potential of poultry industry in reversing the inadequate protein intake by most Nigerians has also been recognized. In comparison to other livestock enterprises, broiler production have the advantage of the fast growth rate, cheaper, high feed conversion efficiency, can be eaten by one family man and is not forbidden by any culture or religion. The Nigerian agricultural economy enjoyed decade of boom in poultry production between mid-70's and mid 80's. For instance, the population of cattle (174.32 million), goat (156.6 million) and sheep (190.31 million) while that of poultry was estimated to be 660 million against other animal population in 1983. The production level was so high that the sub-sectorial economy becomes second in African (Onyebimana, 2000) with proper attention, poultry can be relied upon in a short run solving the deficit in protein supply.

Several suggestions have been offered as a short term measures to combat meat shortages in Nigeria. Mua'dAbdul, *et al.*, (2008) has suggested emphasis on massive poultry production, but the limitations posed by shortages and high cost of food grains and medication were obvious. Even the call for increasing awareness of Nigerians about the serious consequences of animal protein shortages, the encouragement of every Nigerian to keep a few livestock as a hobby by as well as liberalization of credit facilities and subsidies on factors of production for genuine livestock farmers to increase production and reduce price has not significantly solved the problem.

In Benue state, poultry production plays an exceptionally important role; approximately 80% of rural households are engaged in smallholder poultry production (Kryger et al. 2010). Poultry production plays an important role in rural incomes in sub-Saharan Africa; especially in Nigeria (Mengesha 2011; Van der Sluis, 2007). The enforced demand for foods of animal origin could be satisfied especially by the production of poultry, as these products have seen the greatest increase in production in recent years **(**FAO 2011; Speedy, 2003**;** Delgado and Narrod 2002).

Access to credit is expected to enhance farming households' ability to acquire capital intensive technology and assets to facilitate and improve farming activities resulting in greater capacity to invest in cultivation of high yielding crops and larger farm holdings (Emereole, 1995; Nwaru, 2003; Nwaru and Onuoha, 2010; Ammani, 2012). Qureshi *et al*. (1996) observed that an increase in credit to agriculture will lead to increase in food production and farmers' income, because as the demand for credit increases farmers output also increases, resulting in improvement in their wellbeing.

However, agricultural loan remains a critical means through which many problems confronting poultry farmers can be resolved. Primarily, it assists in breaking the chains of the vicious circle of poverty which is the main cause of low productivity and low income of the poultry farmers (Bamiro *et al*., 2012). The level of credit available to these farmers is however, grossly inadequate and therefore, limits the realization of their full potentials. It is against this background that this paper seeks to assess credit acquisition and utilization among small scale poultry farmers in Katsina-Ala and Konshisha local government areas in Benue State, Nigeria.

The objectives of this paper are to:

i. identify sources of agricultural credit among poultry farmers in the study area; and
ii. determine the factors affecting credit acquisition by poultry farmers in the study area;

II. CONCEPT OF AGRICULTURAL FINANCE

According to Ayodele and Adeusi (2002),agricultural finance is all about the acquisition and utilization of capital (i.e. finance), the factor of production that facilitates the acquisition, procurement and management of the other factors of production namely, land, labour, capital – physical, and entrepreneur(management), in agriculture and which, is not only a lubricant but the lifeblood of the economy. It cuts across financial management and the financial institutions serving the agricultural sector of the economy. It is the most important factor in economic development. Capital has two concepts – the physical capital which refers to the physical assets (land, buildings, plants, machinery and equipment) used in the production of goods and services either for further or final consumption, and the finance capital which is used not only to procure the physical assets but also operates and manages the assets on daily basis to ensure continuous production of goods and services.

Finance is seen and viewed as the most important and the most talked about (and still being talked about) problem of

https://dx.doi.org/10.22161/ijeab.51.14 *ISSN: 2456-1878*

agriculture. It is regarded as the greatest limiting factor to the development of agriculture in Nigeria and consequently, the eradication of extreme poverty and hunger. Indeed, World Bank (1975) observed that finance is the only one element in the package of inputs and services needed to raise the productivity of small farmers. Ayodele *et al.*, (2002), see it as one of the basic problems facing the less developed countries is the scarcity of domestic capital relative to the size of investment required to achieve high and self-sustaining rates of growth of national and per capital income.

Finance, in an economy, is basically from two (2) main sources – savings and borrowings. Savings, otherwise regarded as equities, is the basis of money economy which allows the release of production resource for investments in the production of goods and services and which enhances real economic growth. It is that part of the disposable income that is not immediately consumed. Borrowings, on the other hand, are the use of other people's money for investment purposes. While savings (equities) is a direct source of financing in an economy, credit (borrowings) is an indirect source. In the integrated and technology driven economy of today, it is evident that there is no amount of equities that can sustain the expected productivity of agriculture to meet the increasing need of the nation, either individually or corporately. It is therefore apparent that borrowing, otherwise regarded as credits, is the major and most ideal source of adequate financing for agriculture, just like any other commercial venture and/or any sector of the economy. World Bank (2005) recognized that credits constitute the largest component of its agricultural lending. It is the duty of the financial institutions that as the financial intermediaries must intermediate efficiently between the savings unit and the investing unit to sustain continuous availability of borrowings (credits).

The objective of agricultural financing policies is to establish an effective system of sustainable agricultural credit schemes, programmes and institutions that could provide micro and macro credit facilities for small, medium and large scale producers, processors and marketers in the agricultural sector of the economy. The CBN (2005) asserted that "robust economic growth cannot be achieved without putting in place well focused programmes to reduce poverty through empowering the people by increasing their access to factors of production, especially credit."

Study Area

Katsina-Ala Local Government Area (LGA) is one of the twenty three local government areas in Benue State. It was created as a division on February 3, 1976. At the 2006 Census, the LGA had a population of 225,471 persons made up of 114,093 males and 111, 093 females (Federal Government of Nigeria, 2009). Major crop grown include yams, maize, rice, groundnut, beans beniseed and soyabeans. Livestock like piggery, goat rearing and poultry production are commonly found in the local government among other agricultural products.

Konshisha local government area is one of the 23 local government area of Benue State, Nigeria. The Local Government Area has a population of 143,045 (1991 census). Konshisha is essentially an agrarian local government, The people of Konshisha Local Government are predominantly of crops such as yams, cassava, rice, soya beans guinea corn, groundnuts, , oranges, etc. Similarly the people also do raise livestock such as piggery, goat rearing and poultry production.

III. METHODOLOGY

The study used survey design research employing the cross-sectional approach because this entails evaluation of selected respondent views on the subject matter. The study population consists of small scale poultry farmers in Katsina-Ala and Konshisha local government areas in Benue State. To ensure even distribution of the sample for the study, a multistage sampling technique was used in selecting small scale poultry farmers within the study area. In order to arrive at a reasonable sample size, the Yamane, (1973) technique was used. The method is mathematically derived Yamane formula:

$$n = \frac{N}{1 + N(e)^2}$$

Where,

n = required responses/sample size

e^2 = error limit

N = population size

Using the formula at 95% confidence level and an error limit of 5% result in Table 1 below:

Table 1: Sampling Distribution using Bourley's Proportional Allocation Technique

Local governments under study	Population frequency	Sample size distribution using bourley's technique
Katsina-Ala	500	$n_b = \frac{500 \times 350}{2800} = 62$
Konshisha	206	

https://dx.doi.org/10.22161/ijeab.51.14 *ISSN: 2456-1878*

$$n_b = \frac{206 \times 350}{2800} = 26$$

Overall Total	**706**	**88**

Source: Field survey data, 2017

Structured questionnaire was administered to eight –eight (88) randomly selected respondents in the study area and interview technique was also used to obtained additional information from the respondents whenever the need arose. The data collected for the study were analyzed using descriptive and inferential statistical tools.

IV. RESULTS AND DISCUSSION

Available Sources of Agricultural Credit

Table 2 below indicates the available sources of agricultural credit used by small-scale poultry farmers in the study area. These include, Bank of Agriculture (BOA), commercial banks, micro-finance banks, cooperative societies, money lenders, local bam, friends &relatives and personal savings. About 26.8 % and 17.1 % of the respondents received credit from cooperative societies and micro-finance banks respectively. These results could be attributed to the lower bureaucracy and delays associated with requesting and receiving financial loans from cooperative societies as compared to micro-finance banks, and this must have encouraged farmers to borrow more frequently from the cooperative societies. This finding is consistent with those of Hussein (2007); Olagunju (2007); Babalola (2012); Ololade and Olagunju (2013) who posited that loans from credit cooperative may be perceived to be devoid of administrative delays and as such there was no insistence on collateral especially for members of such cooperative societies which motivated members to preferably borrow from their cooperative societies at the expense of micro-finance credit.

The results also revealed that farmers who benefitted from credit facilities from the commercial banks and the Bank of Agriculture (BOA) constituted 6.1 and 6.9% respectively. The low patronage of BOA may also be due to inadequate awareness by the farmers of the existence and availability of such facilities in the rural areas. Furthermore, low patronage of commercial banks may also be attributed to lack or limited presence of these banks in the rural areas, coupled with the delays associated with the approval and disbursement of loans to beneficiaries as well as with the insistence on collateral security from the prospective beneficiaries by the bank authorities. Interest rates of commercial banks are always relatively higher due to the higher transaction cost involved. It could also be insinuated that the setup of commercial banks is usually cumbersome and expensive and exist mostly in urban centres where farmers are less concentrated. This finding agrees with the observations of Okwoche, *et al.*, (2012), who observed that the informal source of credit was more popular among small-scale farmers, and this may be due to the relative ease in obtaining credit devoid of administrative delay, non-existence of security or collateral, and flexibility built into the repayment system, which is against what is obtained in the formal sources.

Table 2: Percentage Distribution of Farmers according to Available Sources of Institutional Credit (n=246)

Sources of credit	Frequency	Percentage
Bank of Agriculture	17	6.9
Commercial Banks	15	6.1
Micro-finance bank	42	17.1
Cooperative Societies	66	26.8
Money lender	35	14.2
Local Bam	33	13 .4
Friends & Relatives	25	10.2
Personal savings	20	8.1

Source: Field survey data, 2017* Multiple responses existed, hence > 100 %

Determinants of Credit Acquisition by Poultry Farmers

The results in Table 3 indicate that five out of the 13 variables in the Logit Model have significant coefficients. These included gender X_2, household size X_3, and amount of credit X_8, main occupation X_9 and access to extension agents X_{13}.

The coefficients of age (X_1) education level (X_4), source of credit (X_5), non -farm income (X_6), net farm income (X_7) and number of dependents (X_{10}) were significant. This implies that age, education level, source of credit, amount of nonfarm

https://dx.doi.org/10.22161/ijeab.51.14 *ISSN: 2456-1878*

income, net farm income and number of dependents of the poultry farmers do not affect the decision on the likelihood of the use of agricultural credit in the study area. The gender of the farmers represented by the coefficient of variable X_2 was positively signed and significant at 10 %. This result, as indicated by the marginal effect, shows that a male farmer has about 90 % likelihood to acquire an agricultural loan to finance a poultry business than a female farmer. This may be attributed to cultural practices of the respondents in the study area as loans are mostly preferably given to males than their female counterparts on the conviction that such loans are more secured in the hands of males than the females. Furthermore, males are more likely to have adequate collateral, such as landed properties and assets that guarantee them access to loans from commercial banks and other formal sources as compared to their female counterparts.

The coefficient of X_3 representing the farmers' household size has negative sign but was significant at 10%. This is in conformity with the a *priori* expectations. Poultry farmers who have large household size are not more likely to use agricultural loans for the purpose for which they were obtained as a result of diverting the loans to solve other emerging family needs, such as clothing, hospital bills, burials and school fees among others. The marginal analysis shows that the likelihood of a poultry farmer to obtain and utilize the agricultural loan will reduce by about 35 % if the household size increases by 1%, thus affected by many other factors that may not have been covered in the present study area.The effects of the amount of loan available to the poultry farmer (X_8) variable on the decision to acquire agricultural credit closely conformed to a priori expectations. The positive coefficient of the parameter estimates for variable X_8 indicates that the more the volume of loanable agricultural credit facilities are provided, the more the likelihood of a decision by poultry farmer to access it. The marginal effect shows that a percentage increase in the volume of loan available to poultry farmers will increase the likelihood of decision to acquire loan by about 12 %.

The negative signs of the coefficients representing poultry farmer's main occupation and access to extension agents indicate contrary results to a priori expectations. As indicated by Table 5, a full-time farmer's likelihood to decide for agricultural loan is about 13 % lower than that of a part-time farmer. Similarly, a poultry farmer with more extension contacts are (about 79 %) less likely to use agricultural loan. This high percentage is as a result of extension training and linkage to other farm input supply sources.

Table 3: Logit Regression Model of decision to use Credit by Poultry Farmers

Variable	**Estimated coefficient**	**Marginal effect**	**Z-value**	**Standard Error**
Constant	-0.1306	-0.5326	-0.225	0.0756
Age (X_1)	0.7546	0.3077	0.323	0.0785
Gender (X_2)	0.2195*	0.8953	1.706*	0.0918
Household size (X_3)	-0.8579***	0.3499	3.659***	0.08596
Education level (X_4)	0.2328	0.9494	1.077	0.13140
Source of Credit (X_5)	0.7332	0.2990	0.383	0.1310
Non Farm Income (X_6)	0.3206	0.2798	1.975	0.9342
Net farm income (X_7)	-0.4489	-0.1831	-0.302	0.2801
Amount of credit (X_8)	0.2861**	0.1166	1.996***	0.1373
Main occupation (X_9)	-0.3200**	-0.1305	-2.015**	0.9342
Number of dependence (X_{10})	0.8092	0.3300	0.656	0.0676
Market Source (X_{11})	-0.3321	-0.1354	-0.565	3310
Credit use experience (X_{12})	-1.3137	-0.5357	-1.325	0.896
Extension service (X_{13})	-1.9316***	-0.7877	-3.189***	0.0270
Log likelihood	-1486.36			
LR Chi2 (10) Proba>chi2 Pseudo R2	42.21 0.000 0.368			

Source: Field survey data, 2017

***, ** and * denote that, the association coefficients are significant at 1, 5 and 10% levels, respectively.

https://dx.doi.org/10.22161/ijeab.51.14 *ISSN: 2456-1878*

V. CONCLUSION

Agricultural credit is adjudged as an important input to increase poultry production. The study shows that certain factors responsible for the decision to obtain or otherwise agricultural loan from any source. This study also shows that poultry enterprise using loans in the study area is profitable; however the profitability level is a function of the scale of production.

VI. RECOMMENDATIONS

The study suggested that government should remove security advancement of collateral conditions that discourages poultry farmers from commercial banks facilities, formal and informal money lenders should reduce the interest rate to one digit for farmers to afford convenient repayment and more borrowers to be encouraged, and lenders should timely approve/disburse fund to farmers for effective utilization and curtail the protocols involved in credit acquisition.

REFERENCES

[1] Abedullah, N., Mahmood, A., Khalid, M. & Kouser, S., (2009). The role of agricultural credit in the growth of livestock sector: A case study of Faisalabad. *Pak. Vet. J., 29: pp81-84*.

[2] Ammani, A. A., (2012). An Investigation into the Relationship between Agricultural Production and Formal Credit Supply in Nigeria. *International Journal of Agriculture and Forestry* 2(1), pp. 46-52

[3] Ayodele, T.D. and Adeusi, S.O.(2002): *Agricultural Financing In Nigeria:*Balfak Educational Publishers, Ado – Ekiti. Nigeria.

[4] Babalola, D. A., (2012). Determinants of Farmers'Adoption of Agricultural Insurance: the Case ofPoultry Farmers in Abeokuta Metropolis of OgunState, Nigeria. *British Journal of Poultry Sciences* 3(2), 36-41.

[5] Babalola, D. A and Babalola, Y., (2013). Economic Effects of Media Campaign against Pandemic diseases: The Case of Bird Flu (H5N1) on poultry business in Ogun state, Nigeria. *Arabian Journal of Business and Management Review* 2(12), 80-88.

[6] Bamiro, O.M., Otunaiya, A.O. & Idowu, A.O. (2012). *Economics of Horizontal Integration in Poultry Industry in South-West Nigeria*. Int. J. Poult. Sci., 11: pp. 39-46.

[7] Central Bank of Nigeria, (2005). Micro-Finance Policy Regulatory and Supervisory Frame Work for Nigeria, CBN, Abuja.

[8] Delgado, C.L. and NarrodC.A. (2002). Impact of Changing Market Forces and Policies on Structural Change in the Livestock Industries of Selected Fast Growing Developing Countries. FPRI.FAO.ttp://www.fao.org/WAIRDOCS/LEAD/X6115E/x6115e00.htm (accessed August 13, 2014).

[9] Emereole, C.O., (1995). Demand for Institutional Credit by Farmers in Abia State, Nigeria: A Case Study of the Nigerian Agricultural and Cooperative Bank, M.Sc. Thesis, Federal University of Technology Owerri, Nigeria.

[10] FAO (2011). Food and Agricultural Organization. Monitoring African Food and Agricultural Policies: Analysis of incentives and disincentives for cassava in Nigeria, Technical notes series, MAFAP, FAO, Rome.

[11] Hussein, H. K, and Ohlmer B. O., (2008). Influence of Credit Constraints on Production Efficiency: The Case of Farm Households in Southern Ethiopia. Swedish University of Agricultural Sciences, Sweden; 2008.

[12] Kryger KN, Thomsen KA, Whyte MA,& Dissing M (2010). Smallholder Poultry Production – Livelihoods, Food Security and Socio-cultural Significance, Smallholder Poultry Production. *FAO.Rome Italy.P. 76.*

[13] Mua'dAbdul, A., Munir, T., Mohsen A. & Abdel, R. A., (2008). Effect of plant Density and Nitrogen rate on Herbage yield of Marjoram under Mediterranean Conditions. *American Eurasian J. Agric and Environmental Sciences*, 3(2): pp.153-158.

[14] Nwaru, J.C. (2003). Gender and relative production efficiency in food crop farming in Abia State, Nigeria. *Nigerian agricultural Journal* 34: pp. 1-30

[15] Nwaru, J.C. and Onuoha, R.E., (2010). Credit Use and Technical Change in Smallholder Food Crop Production in Imo State of Nigeria, *New York Science Journal* 3(11), 144-152

[16] Okonjo, C.N., (2000). The Nigeria poultry industry the past, present and future. A paper presented at the 5th annual conference of Animal society of Nigeria Rivers state University of Science and Technology. Port Harcourt.

[17] Okwoche, V. A, Asogwa, B. C, & Obinne, C. P. O.(2012) Evaluation of agricultural credit utilization by cooperative farmers in Benue State of Nigeria. *European Journal of Economics, Finance and Administrative Sciences.* 47: pp. 123-133.

[18] Olagunju,F. I. (2007). Cost and Returns on egg production in South-Western Nigeria. *Research Journal of Agricultural Economics*, 3(9): pp. 468 – 474.

[19] Ololade, R. A. and Olagunju, F. I., (2013). Determinants of Access to Credit among Rural Farmers in Oyo State, Nigeria, *Global Journal of Science Frontier Research Agriculture and Veterinary Sciences* 13 (2), 50-58

[20] Onyebimana, U.A.U.,(2000). *"Economics and production management for Agriculture* Alphabet Nigeria publisher. Nigeria, pp: 64-71.

[21] Otunaiya, A.O., Okuneye, P.A. &. Aihonsu, J.O. (2012). Pattern of Inorganic Fertilizer use among Food Crop Farmers in Ogun State, Nigeria. *Asian J. Agric. Sci.,4: pp.26-31.Pakistan Development Review*.31:4.

[22] Qureshi, S. K and Shah. A (2002). A Critical Review of Rural Credit Policy in Pakistan. *The Pakistan Development Review*.31:4.

[23] Speedy, A.W. (2003).Global production and consumption of animal source foods. *Journal of Nutrition* 133(11): 4048-4053.

[24] Yamane, T. (1973).*"Statistics: an introduction analysis."* Singapore: Times Printers, c1973.

https://dx.doi.org/10.22161/ijeab.51.15 *ISSN: 2456-1878*

Prebiotic Potential of underutilized Jerusalem artichoke in Human Health: A Comprehensive Review

Diksha Gupta[1*] and Neelam Chaturvedi[2]

[1]Research scholar, [2]Associate professor, Department of Food Science and Nutrition, Banasthali Vidyapith, Dist-Tonk, Rajasthan, India (304022)
E-mail: dikshagupta823@gmail.com

***Abstract*— *The global burden of non-communicable diseases has been rising over the last century, with the leading cause of neurological, metabolic and degenerative disorders. The several studies have reported that the incorporation of prebiotics in human diet is favourable to eliminate the pathological ailments. Since prebiotics occur naturally in plants including leeks, asparagus, onion, wheat, garlic, chicory, oats, soybean and Jerusalem artichoke. Jerusalem artichoke is a perennial tuber contains proteins, mono or poly- unsaturated fatty acids, vitamins, minerals and excellent amount of soluble dietary fibers such as inulin and fructo-oligosaccharides with negligible amount of starch which is digested with Bifidobacterium. It is associated with expansion of bioavailability of minerals, increase activity of favourable bacteria, ease the digestion of high protein diets, delay fat absorption, deliver roughage, prevent constipation, increase satiety value which results in various therapeutic properties such as antidiabetic, cardioprotective and hepatoprotective effects, anti-inflammatory, antimicrobial, anti-obesity, anti-inflammatory and other pharmacological properties. It is also used as a functional food ingredient in the design and production of child formulation, chocolates, sugar confectionaries, soups, sauces, meat products, bakery products, nutritional bars, beverages, milk products, dietary supplements and many other food products. Therefore, its remarkable therapeutic effects and various food applications make this tuber very valuable for further investigation in the area of pharmaceutical and food industries.*

Keywords*— *Jerusalem artichoke, Prebiotics, Inulin, Fructo-oligosaccharides, Pharmacological properties.

I. INTRODUCTION

Nowadays, besides the basic role of nutrition entailing in the supply of nutrients for growth and development, additional aspects are becoming increasingly significant, including the maintenance of health and counteracting diseases. The global burden of non-communicable diseases has been rising over the last century, with the leading cause of neurological, metabolic and degenerative disorders. The several studies have reported that the incorporation of prebiotics in human diet is favourable to eliminate these pathological conditions (Markowiak and Katarzyna, 2017). Prebiotics are non-digestible oligosaccharides and polysaccharides that positively stimulating the growth and/or activity of *bifidobacteria* and lactic acid bacteria in the colon. They exert antagonism against *Salmonella sp.* and *Escherichia coli*, limiting their proliferation, therefore, improve host health (Bindels, *et al.*, 2015). Prebiotics are occurring naturally in plants such as leeks, asparagus, onion, wheat, garlic, chicory, oats, soybean and Jerusalem artichoke and synthesized from enzymatic digestion of polysaccharides. Scientists have re-examined and classified prebiotics on the basis of common criteria in which, Inulin, fructose-oligosaccharides, galactosaccharides, lactulose and polydextose are recognized as the establishing prebiotics. On the other hand, isomalto-oligosaccharides, xylo-oligosaccharides and lactitol are categorized as emerging prebiotics (Sadler and Stowell, 2007). They exert a myriad of health promoting effects including; they are involved in formulating starter culture, maintain intestinal health, inhibiting cancer and preventing metabolic disorders. They also seem to promote a positive modulation of immune system as shown in Figure 1 (Delgado, *et al.*, 2011).

https://dx.doi.org/10.22161/ijeab.51.15 *ISSN: 2456-1878*

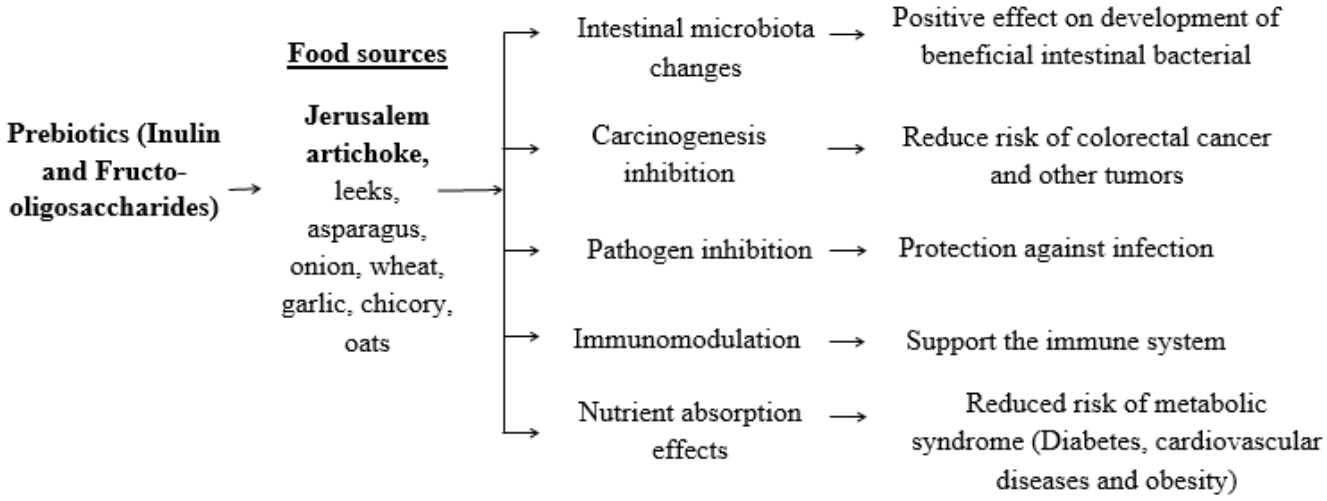


Fig.1: Mechanism of prebiotics and their effects

II. JERUSALEM ARTICHOKE

Jerusalem artichoke, a sunflower species belongs to *Asteraceae* family is botanically known as *Helianthus tuberosus* L. The stem is 5-10 ft tall, ridged and stout which can become woody over time. The leaves are situated near the top of the stem and flowers are small and bright yellow (Pan, *et al.*, 2009). It has an underground rhizome system which bears uneven and elongates varying from knobby to round clusters small fleshy tubers resembling to potatoes. The colour of tubers varies from pale brown, red and purple depending upon the climate conditions (Talipova, 2001). These tubers originated from the United States that become naturalized as an economic crop worldwide in temperate areas and is presently also grown in Canada, France, Germany, Netherlands, USSR, Japan and India (Slimestad, *et al.*, 2010).

Jerusalem artichoke tuber contains proteins, mono or poly-unsaturated fatty acids, vitamins, minerals and dietary fibres with negligible amount of starch (Barta and Patkai, 2007). Its tubers have functional food ingredients such as inulin and fructo-oligosaccharides contributing nutraceutical properties (Kays and Nottingham, 2007). According to Barclay, *et al.*, (2010) that artichoke tubers contain 10-20% of inulin on fresh weight basis and known to have prebiotic effects. Similarly, El-Kholy and Mahrous, (2015) stated that the aqueous extract of Jerusalem artichoke tubers contains higher amount of inulin (21.46g/100g) and three major sugars: sucrose (4.33g/100g), fructose (3.25g/100g) and glucose (2.77g/100g). It has high amount of biologically active components including sesquiterpenes, flavonoids, isoflavonoids, phenols, phenolic acids, glycoalkaloids, phytic acids, coumarins, organic acids, polyacetylenes, and their derivatives naturally occurring isomers of caffeoylquinic acid. It also possesses antidiabetic, anti-inflammatory, antimicrobial, anti-obesity, anticancer and other pharmacological properties (Kapusta, *et al.*, 2013).

III. PREBIOTIC COMPONENTS IN JERUSALEM ARTICHOKE

INULIN

Inulin is a plant polysaccharide that comprises all straight-chain fructans consisting of fructosyl units linked by β-D(2-1) glycosidic bond (Roberfroid, 2005). It is a polydisperse mixture of molecules which can be symbolized by as GFn, where G is the glucosyl moiety, F is the fructosyl moiety and n is the number of fructosyl moiety linked by β(2-1) linkages. The degree of polymerization of inulin typically ranges from 2 to 60 as shown in Figure 2. The presence of β(2-1) bond prevents inulin from being digested like typical carbohydrate and is responsible for its reduced calorie value and dietary fibre effects (Abed, *et al.*, 2016).

Inulin is a “functional food ingredient” and known to have prebiotic potential, which is associated with expanding bioavailability of minerals, inhibition of pathogenic bacteria and increase activity of beneficial bacteria in the digestive tract. It also ease the digestion of high protein diets, deliver roughage, prevent constipation, delay fat absorption, increase satiety value without having extra calories which results in lowering blood glucose, cholesterol and triglycerides levels (Lopez-Molina, *et al.*, 2005).

https://dx.doi.org/10.22161/ijeab.51.15 *ISSN: 2456-1878*

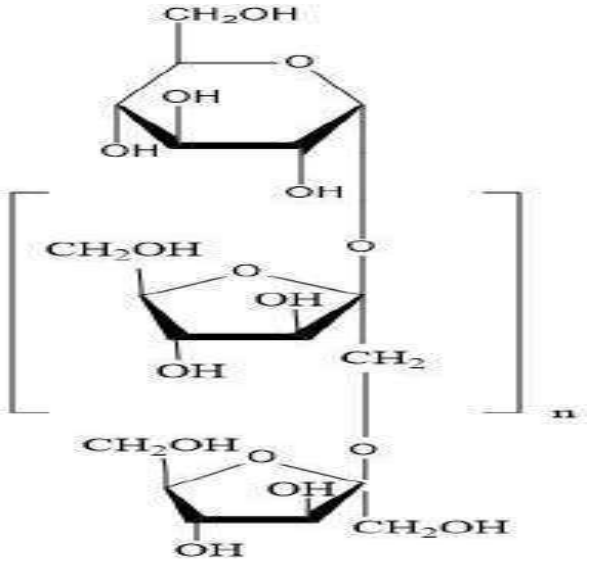


Fig.2: Structure of inulin, n=2-60

FRUCTO-OLIGOSACCHARIDES (FOS)

Fructo-oligosaccharides (FOS), also known as "oligofructan and oligofructose" that are naturally present in vegetables and fruits (Muir, *et al.*, 2009). They are short chain of fructose polymer which are composed of D-fructose units linked with β(2-1) and not hydrolysed by human digestive enzymes. They are obtained from the hydrolysis of inulin using endoinulinase enzyme or by conducting enzymatic reaction of sucrose transfructosylation residues using the β fructofuranosidase or fructosyl- transferase (De-Sousa, *et al.*, 2011). Ketose (GF_2), nystose (GF_3) and fructofuranosyl nystose (GF_4) are the three key chemical structures of FOS, in which the fructose units (F) are linked at β(2-1) glycosidic bonds and the terminal glucose units (G) are linked to fructose unit at the α(1-2) glycosidic bond as shown in Figure 3 (Ibrahin, 2018).

FOS can be used as a substitute for sucrose in foods such as, yogurt, nutritional bars, diet beverages and in low calorie sweetener for diabetes. They are claimed to enhance the growth of favourable bacteria in the colon and used as soluble dietary fibre for constipation and traveler's diarrhoea (Costa, *et al.*, 2015). It is associated with Improving mineral absorption (calcium and magnesium), lowering of blood pressure and responsible for the inhibition of the production of the reductase enzyme that contribute to cancer (Coundray, *et al.*, 2003). It also prevents obesity, stimulates the immune system, reduce the synthesis of triglycerides and fatty acids in the liver and decrease blood glucose levels (Kolida and Gibson, 2007). Table 1 illustrates the amount of inulin and fructo-oligosaccharides present naturally in plants such as Jerusalem artichoke leeks, asparagus, onion, wheat, garlic, chicory, oats and soybean (Thammarutwasik et al., 2009).

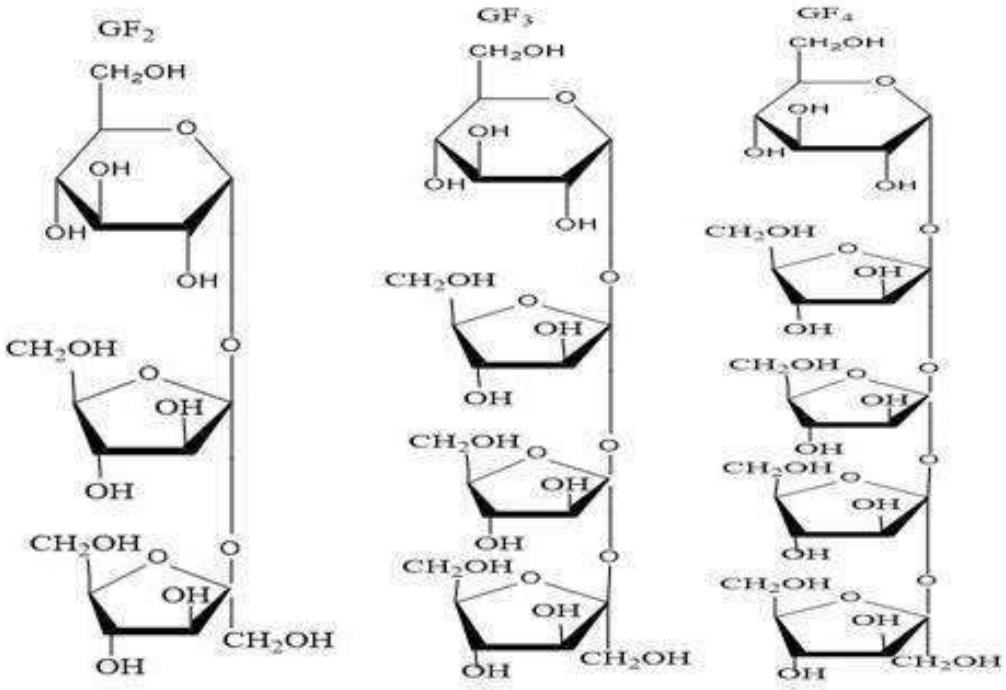


Fig.3: Structure of fructo-oligosaccharides

https://dx.doi.org/10.22161/ijeab.51.15 *ISSN: 2456-1878*

Table 1: Inulin and fructo-oligosaccharides content of food products

Food Sources	Inulin (g/100g)	Fructo-oligosaccharides (g/100g)
Jerusalem artichoke tuber (*Helianthus tuberosus*)	**16.0-20.0**	**10-15**
Raw onion pulp (*Allium cepa*)	1.1-7.5	2.0-6.0
Asparagus raw (*Asparagus officinalis*)	2.0-3.0	5.0-10
Chicory root (*Cichoriumintybus*)	35.7-47.6	5.0-10
Barley (raw cereal) (*Hordeumvulgare*)	0.5-1.5	0.5-1.5
Wheat (flour baked) (*Titicum sp.*)	1.0-3.8	1.0-3.8
Garlic (*Allium sativum*)	9.0-16.0	3.0-6.0
Leek (*Allium ampeloprasum*)	3-10	2-5
Banana (*Musa sapientum*)	0.3-0.7	0.3-0.7
Yacon (*Smallanthus sonchifolius*)	3-19	3-19
Artichoke (*Cynara scolymus*)	3-10	<1
Dandelions (*Taraxacum officinale*)	12-15	NA
Rye (*Secale cereale*)	0.5-1.0	0.5-1.0

FOOD APPLICATIONS OF JERUSALEM ARTICHOKE

Currently, one of the trends in food segment is the health and wellness linked with the growth of food industry. Formulation of functional foods containing prebiotic component is widely used in the design of numerous dietary and pharmaceutical supplements in recent years, not only for economic reasons but by scientific evidence of its remunerations (Burgain, *et al.*, 2011). Hence, Jerusalem artichoke as a source of inulin and fructose-oligosaccharides, have been incorporated into a wide variety of food products. It is used as a functional food ingredient in the design and production of child formulation, chocolates, sugar confectionaries, soups, sauces, nutritional bars, meat products, bakery products, beverages and drinks, yoghurts, desserts, milk products, dietary supplements and many other food products as depicted in Table 2 (Wang, 2009). Though, consumers may appreciate tasty food while promoting beneficial effects to their own health (Coman, *et al.*, 2012). According to Millani, *et al.*, (2009) consumption of child formulations incorporated with these agents is associated with improving allergy cases and preventing constipation. Radovanovic, *et al.*, (2014) conducted that wheat bread fortified with Jerusalem artichoke powder have optimal nutritional value with low Glycemic index (53.70) and low Glycemic load (7.67). Likewise, Rodrigues, *et al.*, (2012) reported that preparation of cheese with inulin and fructo-oligosaccharides is associated with lower atherogenecity index.

Table 2: Application of Jerusalem artichoke in food products

Food Products	Applications
Beverages and drinks	Mouthfeel, sugar replacement, foam stabilization and prebiotics
Yoghurts and desserts	Texture and mouthfeel, sugar replacement, fiber and prebiotics
Meat products	Texture stability, fat replacement, and fiber
Breads and fillings	Texture, sugar or fat replacement, fiber and prebiotics
Cake and biscuits	Moisture retention, sugar replacement, fiber and prebiotics
Dietary supplements	Sugar or fat replacement, fiber and prebiotics
Child formulations	Body and mouthfeel, texture, fiber, stability and prebiotics
Sugar confectionaries	Sugar replacement, fiber and prebiotics
Chocolate	Sugar replacement, heat resistance and fiber
Soups and sauces	Sugar replacement and prebiotics

THERAPEUTIC PROSPECTIVE OF JERUSALEM ARTICHOKE AS FUNCTIONAL FOOD INGREDIENT ANTIDIABETIC PROPERTIES

There is evidence that soluble fibres are beneficial in the reduction of serum glucose and insulin postprandial by

https://dx.doi.org/10.22161/ijeab.51.15 *ISSN: 2456-1878*

raising the viscosity of the nutrients in the small intestine that results in delaying the release of glucose (Saad, 2006). Oral administration of Jerusalem artichoke tuber extracts caused a significant decrease in blood glucose levels by 33.8% in hyperglycemic rats due to the presence of an optimum quantity of polysaccharide inulin (Asian, *et al.*, 2010). Similarly, Al, *et al.*, (2012) elucidated that diets fortified with artichoke tuber induced a significant decrease in serum glucose in the hyperglycemic rats. Wang, *et al.*, (2016) reported that fermented Jerusalem artichoke extract showed significant decrease in blood glucose concentration and serum insulin level in mice. In support of these observations, Okada, *et al.*, (2017) also reported that Jerusalem artichoke tubers improve glucose tolerance in rats. Likewise, Ahn, *et al.*, (2018) revealed that Supplementation of Jerusalem artichoke has been associated with reduced level of fasting glucose and homeostasis model assessment insulin resistance of diabetic patients.

CARDIOPROTECTIVE PROPERTIES

The pronounced decrease occurred in serum total cholesterol, triglycerides, LDL and VLDL in rats fed with Jerusalem artichoke tuber compared with the positive control (Zaky, 2009). Likewise, Gaafar, *et al.*, (2010) stated that supplementation of inulin extracted from artichoke tubers resulted in a decrease in total cholesterol, triglycerides, total lipids, LDL and VLDL-cholesterol levels in diabetic rats. Meanwhile, HDL level was increased significantly. In addition, Asma and Gindy, (2016) have shown that bread substituted with Jerusalem artichoke powder, barley flour and a mixture of both induced significant decrease in triglycerides, total cholesterol and LDL- cholesterol of rats in the hyperglycemic groups in comparison with control group.

HEPATOPROTECTIVE PROPERTIES

The study stated by Ghanem, *et al.*, (2016) concluded that the supplementation of low calorie pan breads containing Jerusalem artichoke as a source of inulin showed significant decrease in Glutamate oxaloacetate transaminase and Glutamate pyruvate transaminase enzyme level in diabetic mice when compared with control group. Kim and Han, (2013) concluded that the aqueous extract of Jerusalem artichoke prevented elevation of aminotransferase, alanine aminotransferase, serum aspartate, y-glutamyl transpeptidase and lactate dehydrogenase levels in STZ-induced diabetic rats. Another result revealed by Yang, *et al.*, (2012), that artichoke tubers may improve hepatic insulin sensitivity, decreases the synthesis of fatty acids and triglycerides in liver and lowers their circulating level in mice. Later, Abdel-Hamid, *et al.*, (2015) concluded that Jerusalem artichoke tubers showed a promising hepatoprotective effect against CCL4 (Carbon tetrachloride)-induced fibrosis via modulation of apoptotic signaling and fibrogenic activity.

ANTIOBESITY PROPERTIES

According to Kaur and Gupta, (2002) that inulin is a low calorie food ingredient as it comprises less than half amount of calorie content of digestible carbohydrates. Cho, *et al.*, (2010) revealed that the supplementation with Jerusalem artichoke tuber exerted the antiobesity effects in the diet of obese rats due to the presence of dietary fibres, rendering a good source for preventing obesity. Later, Guess, *et al.*, (2015) concluded that human subjects supplemented with inulin lost significantly more weight and had lower hepatic muscle fat content compared to control.

ANTI-INFLAMMATORY PROPERTIES

Diets fortified with inulin suppress G protein-coupled receptor-43 overexpression which combat high-fat-diet-induced obesity through the modification of the gut microbiota and resulted in decreased level of circulating lipopolysaccharide and lower C-reactive protein levels to attenuate inflammation (Dewulf, *et al.*, 2013). According to Koleva *et al.*, (2012) administration of inulin and fructo-oligosaccharides attenuate chronic intestinal inflammation in HLA-B27 transgenic rats. Hence, administration of Jerusalem artichoke might reduce systemic inflammation due to the presence of fructo-oligosaccharides and inulin.

ANTIMICROBIAL PROPERTIES

The administration of artichoke as a source of soluble fibers increase the number of *lactobacilli*, *bifidobacteria*, and certain butyrate-producing bacteria such as *Clostridium perfringens* group (Costabile, *et al.*, 2010). Likewise, the study showed that its extracts exerted antifungal activity against *Rhizoctoniasolani*, *Botrytis cinerea* and *Alternariasolani* (Liu, *et al.*, 2007). Gengaihi, *et al.*, (2009) reported that tuber extracts have antimicrobial activity against the test gram-positive (*Candida albo, Pseudomonas, Bacillus subtilis* and *Staphylococcus aureus*) and gram-negative bacteria (*Saccharomyces cerevisiae, Arthrobacter, Kill bacteria tiffy, Escherichia coli,* and *Enterobacter* due to the presence of many potent compounds such as bitter sesquiterpene, lactones, inulin, flavonoids and coumarins.

IV. CONCLUSION

The review paper revealed that Jerusalem artichoke tuber as prebiotic agent appears to be unique among the currently available adaptogenic tubers. It possesses antidiabetic, cardioprotective and hepatoprotective effects, antiobesity, anti-inflammatory, antimicrobial and many

other pharmacological properties. Its powder has been used in numerous dietary supplements such as child formulation, chocolates, sugar confectionaries, soups, sauces, nutritional bars, meat products, bakery products, beverages, milk products and many other products in food industry. Hence, its remarkable therapeutic effects and various food applications make this tuber very valuable for further investigation in the area of pharmaceutical and food industries.

REFERENCES

[1] Abdel- Hamid N, Nazmy MH, Wahid A, Abdel-Moniem M. Jerusalem artichoke attenuates experimental hepatic fibrosis via modulation of apoptotic signalling and fibrogenic activity. Biochem Biotechnol Res 2015; 3(3):43-50.

[2] Abed S, Ali AH, Nowan A, Sobianiazi, Al-Fargaammar, Bakry AM. Inulin as prebiotics and its applications in food industry and human health: A review. Int J Agric Innov Res 2016; 5(1).

[3] Ahn HY, Kim M, Seo CR, Yoo HJ, Lee SH, Lee JH. The effects of Jerusalem artichoke and fermented soyabean powder mixture supplementation on blood glucose and oxidative stress in subjects with prediabetes or newly diagnosed type 2 diabetes. Nutr Diabetes 2008; 8(1):1-13.

[4] Al SH, Sarwat MI, El-Tobgy KM, Abd EI, Alem MS. Jerusalem artichoke tubers, onion and hulless barley as functional foods in alloxan Induced diabetic rats. J Appl Sci Res 2012; 8(2):1328-1336.

[5] Asian M, Orhan DD, Ergun F. Hypoglycemic activity and antioxidant potential of some medicinal plants traditionally used in turkey for diabetes. J Ethanopharmocol 2010; 128(2):384-389.

[6] Asma A, Gindy EI. (2016). Technological and biological studies on Jerusalem artichoke and barley. Eur J Food Sci Technol 2016; 4:1-15.

[7] Barclay TG, Ginic-Markovoic M, Cooper PD, Petrovsky N. Inulin: A versatile polysaccharide with multiple pharmaceutical and food chemical uses. J Excip Food Chem 2010; 1(3):27-50.

[8] Barta J, Patkai G. Chemical composition and storability of Jerusalem artichoke tubers. Acta Aliment 2007; 36(2):257-267.

[9] Bindels LB, Delzenne NM, Cani PD, Walter J. Towards a more comprehensive concept for prebiotics. Nat Rev Gastro Hepat 2015; 12(5):303-310.

[10] Burgain J, Gaiani C, Linder M, Scher J. Encapsulation of probiotic living cells: From laboratory scale to industrial applications. J Food Eng 2011; 104(4):467-483.

[11] Cho KD, Kim EJ, Kim MY, Kim JS, Han CK, Lee BH. Antiobesity and antidiabetic effects of Jerusalem artichoke and purple sweet potato in the diet induced obese rats. Nutrition 2010; 24:722-723.

[12] Coman MM, Cecchini C, Verdenelli MC, Silvi S, Orpianesi C, Cresci A. Functional foods as carriers for SYNBIO®, a probiotic bacteria combination. Int J Food Microbiol 2012; 157(3):346-352.

[13] Costa GT, Abreu GC, Guimaraes AB, Vasconcelos PR, Guimaraes SB. Fructo-oligosaccharide effects on serum cholesterol levels: An overview. Acta Cir Bras 2015; 30(5):366-370.

[14] Costabile A, Kolida S, Klinder A, Gisti E, Baueriein M, Frohbrug C, et al. A double blind, placebo-controlled, cross over study to establish the bifidogenic effect of a very long chain inulin extracted from globe artichoke (Cynara scolymus) in healthy subjects. Br J Nutr 2010; 104:1007-1017.

[15] Coundray C, Demigne C, Rayssiguier Y. Effects of dietary fiber on magnesium absorption in animals and humans. J Nutr 2003; 133(1):1-4.

[16] Delgado GTC, Tamashiro WMSC, Junior MRM, Moreno YMF, Pastore GM. The putative effects of prebiotics as immunomodulatory agents. Food Res Int 2011; 44(10):3167-3173.

[17] De-Sousa VMC, Dos-Santos EF, Sgarbieri VC. The importance of prebiotics in functional foods and clinical practice. Food Nutr Sci 2011; 2:133-144.

[18] Dewulf EM, Cani PD, Claus SP, Fuentes S, Puylaert PG, Neyrinck AM, et al. Insight into the prebiotic concept: Lessons from an exploratory, double blind intervention study with inulin-type fructans in obese women. Gut 2013; 62(8):1112-1121.

[19] El-Kholy WM, Mahrous H. Biological studies on bio-yoghurt fortified with prebiotics obtained from Jerusalem artichoke. Food Nutr Sci 2015; 6(16):1552-1564.

[20] Gaafar AM, Serag EI-Din MF, Boudy EA, EI-Gazar HH. Extraction condition of inulin from Jerusalem artichoke tubers and its effects on blood glucose and lipid profile in diabetic rats. J Am Sci 2010; 6(5):36-43.

[21] Gengaihi SAEI, Aboul Enein AM, Abou Elalla FM, Abou Baker DH. Molecualr characterization and antimicrobial activities of chicory and Jerusalem artichoke plants. Int J Acad Res 2009; 1(2):66-71.

[22] Ghanem SM, Ibrahium MI, Saed B. Effect of low calorie pan bread containing wheat bran and some vegetables flours on biological properties of diabetic rats. Middle East J Appl Sci 2016; 6:315-328.

[23] Guess ND, Dornhorst A, Oliver N, Bell JD, Thomas EL, Frost GS. A randomized control trial: the effect of inulin on weight management and ectopic fat in subjects with prediabetics. Nutr Metab 2015; 12(36):2-10.

[24] Ibrahim OO. Functional oligosaccharides: Chemicals structure, manufacturing, health benefits, applications and regulations. J Food Chem Nanotechnol 2018; 4(4):65-76.

[25] Kapusta I, Krok ES, Jamro DB, Cebulak T, Kaszuba J, Salach RT. Identification and quantification of phenolic compounds from Jerusalem artichoke (*Helianthus tuberosus* L.) tubers. J Food, Agric, Environ 2013; 11:601-606.

[26] Kaur N, Gupta AK. Application of inulin and oligofructose in health and nutrition. J Biosci 2002; 27(7):703-714.

[27] Kays SJ, Nottingham SF. Biology and chemistry of Jerusalem artichoke: *Helianthus tuberosus* L. Florida: CRC Press Boca Raton; 2007.
[28] Kim HS, Han GD. Hypoglycemic and hepatoprotective effects of Jerusalem artichoke extracts on streptozotocin-induced diabetic rats. Food Sci Biotechnol 2013; 22(4):1121-1124.
[29] Koleva PT, Valcheva RS, Sun X, Ganzle MG, Dieleman LA. Inulin and fructo-oligosaccharides have divergent effects on colitis and commensal microbiota in HLA-B27 transgenic rats. *Br J Nutr* 2012; 108(9):1-11.
[30] Kolida S, Gibson GR. Prebiotic capacity of inulin type fructans. J Nutr 2007; 137(11):2503-2506.
[31] Liu HW, Liu ZP, Liu L, Zhao GM. Studies on the antifungal and chemical components of extracts from *Helianthus tuberosus* leaves. J Nat Prod Res Dev 2007; 19:405-409.
[32] Lopez-Molina D, Navarro-Martinez MD, Rojas-Melqarejo F, Hiner AN, Chazarra S, Rodrique-Lopez JN. Molecular properties and prebiotic effect of inulin obtained from artichoke (*Cynara scolymus* L.). Phytochemistry 2005; 66(12):1448-1476.
[33] Markowiak P, Slizewska K. Effects of probiotics, prebiotics, and synbiotics on human health. Nutrients 2017; 9(9):1-30.
[34] Millani E, Konstantyner T, Taddei JAAC. Effects of prebiotics (oligosaccharides) use on child's health. Rev Paul Pediatr 2009; 27(4):436-446.
[35] Muir JG, Rose R, Rosella O, Liels K, Barrett JS, Shepherd SJ, Gibson PR. Measurement of short chain carbohydrates in common Australian vegetables and fruits by high-performance chromatography (HPLC). J Agric Food Chem 2009; 57(2):554-565.
[36] Okada N, Kobayashi S, Moriyama K, Miyataka K, Abe S, Sato C, Kawazoe. *Helianthus tuberosus* (Jerusalem artichoke) tubers improve glucose tolerance and hepatic lipid profile in rats fed a high fat diet. Asia Pac J Trop Med 2017; 10(5):439-443.
[37] Pan L, Sinden MR, Kennedy AH, Chai H, Linda E, Watson LE, Graham TL, Kinghorn AD. Bioactive constituents of *Helianthus tuberosus* (Jerusalem artichoke). Phytochem Lett 2009; 2(1):15–18.
[38] Radovanovic AM, Milovanovic OZ, Kipic MZ, Ninkovic MB, Cupara SM. Characterization of bread enriched with Jerusalem artichoke powder content. J Food Nutr Res 2014; 2(12):895-898.
[39] Roberfroid M. Introducing inulin-type fructan. Br J Nutr 2005; 93(1):13-25.
[40] Rodrigues D, Rocha-Santos TAP, Gomes AM, Goodfellow BJ, Freitas AC. Lipolysis in probiotic and symbiotic cheese: The influence of probiotic bacteria, prebiotic compounds and ripening time on free fatty acid profiles. Food Chem 2012; 131(4):1414-1421.
[41] Saad SMI. Probiotics and prebiotics: The state of the art. Braz J Pharm Sci 2006; 42(1):1-16.
[42] Sadler M, Stowell JD. Calorie control and weight management. In K O'Donnell, MW Kearsley. Sweeteners and sugar alternatives in food technology. 2nd ed. Blackwell Publishing Ltd; 2007. P. 77-88.
[43] Slimestad R, Seljaasen R, Meijer K, Skar SL. Norwegian-grown Jerusalem artichoke (*Helianthus tuberosus* L.): Morphology and content of sugars and fructo-oligosaccharides in stems and tubers. J Sci Food Agric 2010; 90(6):956-964.
[44] Talipova M. Lipids of *Helianthus tuberosus* L. Chem Nat Compd 2001; 37(3):213–215.
[45] Thammarutwasik P, Hongpattarakere T, Chantachum S, Kijroongrojana K, Itharat A, Reanmongkol W, Tewtraku S, Ooraikul B. Prebiotics – A review. Songklanakarin J Sci Technol 2009; 31(4):401-408.
[46] Wang Y. (2009). Prebiotics: Present and future in food science and technology. Food Res Intl 2009; 42(1):8-12.
[47] Wang Z, Hwang SH, Lee SY, Lim SS. Fermentation of purple Jerusalem artichoke extract to improve the α glucosidase inhibitory effect in vitro and ameliorate blood glucose in db/db mice. Nutr Res Pract 2016; 10(3):282-287.
[48] Yang HJ, Kwon DY, Kim MJ, Kang S, Kim DS, Park S. Jerusalem artichoke and chung cooking and additively improve insulin secretion and sensitivity in diabetic rats. Nutr Metab 2012; 9(112):1-12.
[49] Zaky EA. Physiological response to diets fortified with Jerusalem artichoke tubers (*Helianthus tuberosus* L.) powder by Diabetic rats. Am Eurasian J Agric Environ Sci 2009; 5:682-688.

https://dx.doi.org/10.22161/ijeab.51.16 ISSN: 2456-1878

Blood Analysis of Growing Rabbits Fed Cooked Bambara Nut Meal as Replacement for Groundnut Cake in a Semi-Arid Zone of Nigeria

Usman, Y.[1]; Husa, H.[2]; Yusuf, S. Z.[3]; Bukar, B. S.[4]; Dunya, A. M.[5]

[1] Dept. of Agric. Education, Kashim Ibrahim College of Education Maiduguri, Nigeria
[2] Dept. of Animal Production Technology, Ramat Polytechnic Maiduguri, Nigeria
[3]Dept. of Animal Science and Range Management, Modibbo Adama University of Technology, Yola, Nigeria
4 Dept. of Immunology, University of Maiduguri Teaching Hospital, Maiduguri, Nigeria
5 Dept. of Animal Science, Federal University of Gashua, Yobe State, Nigeria

Abstract**—** *A ten-weeks feeding trial was conducted to determine the effect of replacing groundnut cake (GNC) with cooked Bambara nut meal (CBNM) on the Heamatology and serum biochemistry of Growing Rabbits. Thirty mixed breed (New Zealand White X Dutch) of rabbits six to seven weeks of age with initial body weight of 604.50 g were caged individually and allotted to 5 dietary treatments. Each treatment had six (6) replications. The diets and clean drinking water were given ad libitum throughout the period of experiment. In diets 1(control), 2, 3, 4 and 5 CBNM replaced GNC at 0%, 25%, 50%, 75% and 100%, respectively. All data collected in the course of the experiment were subjected to analysis of variance (ANOVA) using the completely randomized design (Steel and Torrie,1980) and where applicable Duncan's multiple range test, was used for meanseparation. The haematological parameters indicated that there were no significant (P>0.05) differences among treatment groups for PCV, RBC, WBC, MCV, MCH and MCHC, while Hb differ significantly (P<0.05) with animals on treatment 5 (100%CBNM) having highest value of 12.73 g/dl while animals on the control (0%CBNM) had 11.10 g/dl as the lowest. There were no significant differences (P>0.05) among treatment groups in total protein, albumin, creatinine, urea, cholesterol and glucose. Globulin range of 4.67-11.00 g/l was obtained with significant difference (P<0.05) among treatment groups with T3 (50%CBNM) having higher value of 11.00 g/l and T4 (75%CBNM) having 4.67 g/l the lowest. Serum electrolytes; Phosphorus (P), Calcium (Ca) and bicarbonate showed no significant (P>0.05) difference among treatment groups.*

Keywords**— Heamatological, Biochemical, Growing Rabbits and Cooked Bambara Nut Meal.**

I. INTRODUCTION

There is a widespread malnutrition in developing countries like Nigeria due to lack of animal protein consumption. The present state of the economy i.e recession coupled with current inflationary trend, scarcity and cost of production contribute to the poor production of animal protein which is required in high demand. FAO (1990), reported that out of the 44g protein supply *per caput*, animal products constitute about 2%, leading to malnutrition and under nutrition of all ages in Nigeria. To overcome the animal protein insufficiency, the need to improve on the feeding management and productive performance of livestock in Nigeria become imperative. This can be achieved by the introduction of the non-traditional meat sources such as rabbits for small scale farmers to rear for its meat and other by-products. Rabbit production and consumption in Nigeria as a livestock species is fast gaining importance and popularity in the semi-arid zone. The have a high feed conversion ratio and are efficient converters of plants products and seeds (Asuquo, 1997). The meat is richer in protein, certain vitamins and minerals, low in fats, and nutritious with high calorie value compared to meat of other species. The high cost of conventional feed ingredients as a result of competition between human and livestock brings about persistent shortfall of animal protein in the diets of most people living in developed countries, which invariably leads to undernourishment. The use of non-conventional feed such as Bambara groundnut to feed fast-growing animals like rabbit should be given attention. The study will serve as a step towards improving the nutritional status and economic well-being of peasant farmers in Maiduguri.

https://dx.doi.org/10.22161/ijeab.51.16 *ISSN: 2456-1878*

II. MATERIALS AND METHODS

The feeding trial was conducted at the Ramat Polytechnic Teaching and Research, Maiduguri. Maiduguri is located within latitude 11^0 5'north and longitude 13^0 9′east (Encarta, 2007). It has an altitude of 354m above sea level (Alaku, 1983). The vegetative zone falls within the sahelian region of West Africa. The annual rainfall varies from 500-600mm with short duration of 3-4 months rainy season; long dry season of 7-8 months is prevalent. According to Ugherughe and Ekedolum (1986), the mean relative humidity ranges from 30%-50% around February to March, while maximum record of 90% is observed around August. Ambient temperatures are higher during the months of April to May and may reach up to 40^0 C and above (Alaku, 1983). According to Aliyu (2007), ambient temperature could be as low as 20^0C during the cold season while during the hot period, which is between February to June, it can reach 44^0 C.

Method of Processing Bambara groundnut seeds

The Bambara groundnut seeds were subjected to cooking at boiling point (100^0C) for a period of one hour in an aluminum cooking pot containing water sufficient enough to cover the seeds using firewood as a source of fuel. After cooking for the period of one hour, the seeds were separated from the water and sun-dried for five days. This is to ensure complete reduction of moisture for ease of milling as corroborated by Omoikhoje *et al*. (2005) and Omoikhoje *et al*. (2006). The sun-dried seeds were then milled and used for the preparation of the experimental diets.

Experimental stock and management

Thirty (30) mixed breed (New Zealand white X Dutch) of rabbits 6-7 weeks of age, were used for the feeding trial that lasted for the duration of ten (10) weeks, excluding one (1) week of adjustment period. The rabbits were weighed and randomly assigned to five (5) different dietary treatments, each treatment containing six (6) replicates. The rabbits were kept in separate cages made from wire with dimensions of 42cm x 42cm x 43cm (LX W X H). Cages were raised above the ground level for ease of cleaning. Metallic feeding trough and plastic drinkers were provided in each cage. Water and feed were provided *ad libitum* throughout the period of experiment.

Experimental diets

Five (5) experimental diets were prepared in which cooked Bambara groundnut meal (CBGM) replaced groundnut cake (GNC) as a source of protein at 0%, 25%, 50%, 75% and 100% levels in diets 1,2,3,4 and 5 respectively that produced is nitrogenous and is calorie diets formulated to contain 18% crude protein and 3437 metabolizable energy (kcal/kg).

The parameters measured

The parameters measured were haematological and biochemical characteristics. At the end of the 7^{th} week of the experiment, blood samples were collected from 3 rabbits per treatment for haematolgical investigation. Blood sample was collected via ear veins of sampled rabbits. Disposable syringe and needle of 20 mm gauges was used. The rabbits were fasted overnight (12 hours) and bled in the morning to collect blood. Fasting was to avoid temporary elevation of blood metabolites as a result of feeding (Bush, 1975). Sterilization of collection site was carried out using alcohol, while xylenes was applied to dilate the veins. After collection of blood samples, sterile cotton wool was applied to cover the pierced veins. Collected blood samples were emptied into bottles containing dipotassium salt of ethylene diamine tetra-acetic acid. (EDTA-K^{2+}) as anti-coagulant, while blood to be used for serum analysis were collected in separate bottles without anti-coagulant. The blood samples were analysed according to Bush (1975) in the Haematology Laboratory, University of Maiduguri Teaching Hospital (UMTH). Collected blood samples were emptied into plain bottles and allowed to stand for coagulation at room temperature. The samples were then centrifuged for ten minutes at 2,000 revolutions per minutes (rpm) to separate the cell from serum.The total protein, albumin and globulin in the serum were analysed using Sigma assay kits (Sigma Chemical Co. St. Louis, Missouri, USA). The total serum protein and serum albumin were determined by Biuret reactions (Bush, 1975). The total serum protein was first estimated and then performing fractionation on further volume of the sample to precipitate and remove globulins; this leaves only albumin in solution.The serum urea estimation was carried out by the Diacetly Monoxime (WHO, 1980). Here, the protein was first precipitated by trichloroacetic acid. The urea in the filtrate then reacted with diacetyl monoxime in the presence of acid, oxidizing reagent and thiosemicarbazide to give a coloured solution. This was then measured in a photoelectric colorimeter at a wavelength of 520 nm.

$$\text{Urea Concentration (mmol/l)} = \frac{AT}{AR} \times 100$$

Where:

AT = Absorbance of test sample

AR= Absorbance of reference sample

Serum Cholesterol was determined by colorimeteric enzyme method as outlined by Bush (1975). The method involves enzymatic hydrolysis and oxidation which terminates in the production of a red coloured solution. The concentration was determined after reading the colorimeter at 546 nm. The serum glucose was estimated

https://dx.doi.org/10.22161/ijeab.51.16 *ISSN: 2456-1878*

by orthotoliudine method as described in WHO (1980). In this method, protein was first precipitated by tricholoroacetic acid. The glucose in the filtrate reacted with orthotoluidine reagent to give a green colour; this was then measured in a photoelectric colorimeter at a wavelength of 630 nm.

Concentration of glucose = $\frac{AT}{AR}$ X 200 (mmol/l)

Where:

AT = Absorbance of test sample

AR= Absorbance of reference sample

Data collection

All data collected in the course of the experiment were subjected to analysis of variance (ANOVA) using the randomized complete block design (Steel and Torrie, 1980) and where applicable, Duncan's multiple range test (Duncan, 1955) was used for mean separation.

III. RESULTS AND DISCUSSION

The haematological parameters are shown in Table 1. There were no significant (P> 0.05) difference among the treatments in their packed cell volume which ranges between 35.33 and 41.00%. The control group (0%CBGM), however, has lower PCV of 35.33%. The PCV range obtained in this study were within the normal range of 31% to 50% recommended by Canadian Council on Animal Care (1984) and closer to the range of 50% reported by Medirabbit (2003) as the ideal for healthy rabbits. Ehebha *etal*. (2008) reported PCV range of 32.50% to 39.69% when they fed CBGM to weaner rabbits and described it as adequate for growing rabbits receiving proper nourishment. There were significant differences (P<0.05) in the Hb values among rabbits receiving the various treatments diets. The range of 11.10 g/dl to 12.73 g/dl which was observed in this experiment is within the range of 9.40 g/dl to 17.40 g/dl reported in Medirabbit (2003) and closer to values of 10.50 g/dl to 12.95 g/dl obtained by Ehebha *et al*. (2008) who fed CBGM to growing rabbits. The values of Hb obtained in this study are within the normal range for adequate metabolism and as such there is no sign of aneamia in the rabbits.There were no significant differences (P>0.05) among the treatments in the RBC and WBC. The range for the RBC observed was 4.10-4.93 X 10^6 µl which is comparable to the values of 3.85-4.66 X 10^6 µl and 3.55-3.81 X 10^6 µl reported by Ehebha *et al.* (2008) and Omoikhoje *et al*. (2004) respectively for young rabbits. Therefore, the values obtained in this study could be considered as adequate for growing rabbits thus, indicating that the rabbits are in a good condition of health with normal functioning of body systems. The WBC range observed in this experiment was 7.23-8.27 X 10^3 µl. There was no significant difference (P>0.05) among treatment groups and the range conformed with the value (7.89 X 10^3 µl) reported earlier by Schalm *et al*. (1975) and within the recommended range of 5 to 13 X 10^3 µl in Medirabbit (2003). The values of WBC obtained indicated that the rabbits were in good health condition and in a state of readiness to combat attack by foreign bodies as indicated by Ehebha *et al*. (2008). There was no significant (P>0.05)difference among treatment groups with respect to the MCV, MCH and MCHC. The MCV range of 66.10 fl to 73.37fl compares favourably with the values of 50 fl to 75 fl reported in Medirabbit (2003) and 68.20 fl ± 4.10 fl by Schalm *et al.* (1975). The MCH range of 20.33 Pg to 24.27 Pg and MCHC of 30.27% to 31.23 % observed in this experiment were close to the values of 18 to 24 Pg and 27% to 34% for MCV and MCHC respectively reported in Medirabbit (2003). Since there was no indication of anaemia among rabbits, the treatment diets are nutritionally adequate for growing rabbits and could support good health and normal growth. The following serum were investigated: total protein, albumin, globulin, creatinine, urea, glucose, calcium, phosphorus and bicarbonate. The results obtained for the biochemical indices are presented in Table 1. There was no significant difference (P>0.05) among treatment groups in the total protein and albumin. Total protein value of 62.00 g/l to 67.67 g/l obtained were within the reference values of 50 g/l to 75 g/l reported in Medirabbit (2003). Albumin value range of 52.00 g/l to 62.33 g/l obtained was higher than the reference value of 25 g/l to 40 g/l reported in Medirabbit (2003) for albumin. However, high serum protein and high albumin values are indications of good protein quality according to Omoikhoje *et al*. (2004). There were significant difference (P<0.05) among treatment groups in the globulin values obtained. The globulin value of 4.67g/l to 11.00 g/l were lower than values of 20.94 g/l to 28.11 g/l and that of 25 g/l to 40 g/l reported by Ehebha *et al*. (2008) and Medirabbit (2003) respectively. However, low globulin could be due to high albumin as the two are inversely related. There were no significant differences (P>0.05) among treatments groups in the values of creatinine recorded. The values of 98.67-121.67 µmol/l observed for creatinine were within the range of 53-124(µmol/l) reported in Medirabbit (2003). The values for urea recorded (3.20 mmol/l to 8.03 mmol/l) were lower than 9.1 mmol/l to 25.5 mmol/l reported in Medirabbit (2003) but closer to the values of 4.20 mmol/l to 6.80 mmol/l reported by Archetti *et al*. (2008). The values of the urea and other indices such as cholesterol, glucose and serum electrolytes (phosphorus, calcium and bicarbonate)

https://dx.doi.org/10.22161/ijeab.51.16 *ISSN: 2456-1878*

were similar in all the treatments. The cholesterol values of 2.23 mmol/l to 5.27 mmol/l were higher than 0.10 mmol/l to 2.00 mmol/l reported in Medirabbit (2003). The higher cholesterol could be due to higher fat content of Bambara nut as indicated by proximate analysis (8.13%) which was closer to the values of 7.15% and 8.31% for raw and roasted Bambara nut reported by Akande *et.al.* (2009). Omoikhoje *et al.* (2004) had earlier explained that cooked Bambara groundnut meal does not contain cholesterol levels that can pose health hazard. The glucose ranges of 5.80 to 7.00 (mmol/l) were within the range of 4.20 to 8.90 (mmol/l) reported in Medirabbit (2003) as normal for growing rabbits. The serum electrolytes (phosphorus, calcium and bicarbonate) had ranges of 2.4. mmol/l to 2.56 mmol/l, 0.73 mmol/l to 0.96 mmol/l and 20.67 mmol/l to 21.33 mmol/l respectively in this experiment. This is an indication that the nerve functions and nerve disposition of the animals are normal.

IV. CONCLUSION AND RECOMMENDATION

The continued increase in the demand for animal protein in the diet of people living in developing countries calls for an increase in production of fast-growing and prolific animals such as rabbit using non-conventional feed ingredients like Bambara groundnut.

In this study cooked Bambara groundnut meal was found to be suitable for the feeding of growing rabbits at different levels of inclusions, from 25% to 100% as a replacement for groundnut cake, a conventional plant protein source. Rabbits fed varied levels of CBGM have shown good performance in terms of weight gain, feed efficiency and nutrient digestibility. The diet was also found to have no deleterious effects on growing rabbits as revealed by the haematological and serum biochemical indices of the rabbits fed the various diets. Although CBGM can replace 100% of the GNC in the rabbit's diets, the inclusion of up to 50% of CBGM in the diet of growing rabbits as replacement for GNC gave optimum economic benefits. The use of CBGM in the feeding of growing rabbits is a simple and cheaper method of overcoming the adverse effects of the anti-nutritional factors. From this experiment it can be concluded that cooked Bambara groundnut meal (CBGM) can replace groundnut cake (GNC) at different levels of inclusions but 50% CBGM diets should be used for optimum economic benefit. However, to obtain more information, it is recommended that investigations be extended to cover other age groups and classes of rabbits such as fattening, pregnant and lactating rabbits.

Table1: Hematological indices of rabbits fed varied levels of cooked bambara groundnut meal (CBGM) as replacement for groundnut cake (GNC)

	Levels of GNC replaced by CBGM					
Parameters	T1 (0%)	T2 (25%)	T3 (50%)	T4 (75%)	T5 (100%)	SEM
Packed cell volume (%)	35.33	41.00	38.00	38.00	41.00	0.0137^{NS}
Haemoglobin concentration (g/dl)	11.10^{c}	12.43^{bc}	11.63^{ab}	11.80^{ab}	12.73^{a}	0.2689*
Red blood cells (x 10^6 µl)	4.20	4.13	4.93	4.27	4.10	0.2994^{NS}
White blood cells (x10^3 µl)	8.10	7.23	7.53	8.27	7.50	1. 3019 ^{NS}
Mean corpuscular volume (fl)	73.37	66.10	70.77	69.53	72.30	4.0849 ^{NS}
Mean corpuscular haemoglobin (pg)	24.27	20.33	21.87	21.63	22.60	1.259 9^{NS}
Mean corpuscular haemoglobin conc. (%)	30.27	30.77	30.99	31.17	31.23	0.4621 ^{NS}

SEM= standard error of means
NS= not significant (P>0.05)
* = Significant difference (P>0.05)

https://dx.doi.org/10.22161/ijeab.51.16 *ISSN: 2456-1878*

a, b, c. = means in the same row bearing different superscripts differ significantly (P<0.05).

Table 2: Serum biochemical indices of rabbits fed varied levels of cooked Bambara groundnut meal (CGBM) as replacement for groundnut cake (GNC)

Parameters	Levels of GNC replaced by CBGM T1 (0%)	T2 (25%)	T3 (50%)	T4 (75%)	T5 (100%)	SEM
Total protein (g/l)	62.33	67.67	65.33	62.00	68.00	3.0614 ^{NS}
Albumin (g/l)	52.00	57.33	54.33	57.33	62.33	3.1894 ^{NS}
Globulin (g/l)	10.33^{b}	10.34^{b}	11.00^{a}	4.67^{c}	5.67^{c}	1.0775*
Creatinine (µmol/l)	121.67	118.00	115.33	98.67	108.33	12.212 ^{NS}
Urea (mmol/l)	5.93	8.03	3.20	7.00	6.13	0.6559^{NS}
Cholesterol(mmol/l)	3.57^{ab}	3.20^{ab}	5.27^{a}	4.53^{ab}	2.23^{b}	0.7464*
Glucose (mmol/l)	5.80	6.01	7.00	6.20	5.83	0.3889^{NS}
Calcium (mmol/l)	2.56	2.50	2.40	2.40	2.50	0.0830 ^{NS}
Phosphate (mmol/l)	0.73	0.96	0.76	0.90	0.80	0.1160 ^{NS}
Bicarbonate(mmol/l)	20.67	21.33	20.67	21.00	20.00	0.5055 ^{NS}

SEM = standard error of means

NS = not significant (P>0.05)

* Significant difference (P>0.05)

a, b, c = means in the same row bearing different superscripts differ significantly (P<0.05).

REFERENCES

[1] Akande, K.E., Abubakar, M.M., Adegbola, T.A., Bogoro, S.E., Doma, U.D. and Fabiyi, E.F. (2009). Nutrient composition and uses of Bambara groundnut (*Voandzeiasubsterranea* (L) VERDCOURT). *Continental. J. Food Sci. Technol*. 3:8-13. Wilolud online journals, 2009.

[2] Alaku, S.O. (1983). Body and carcass losses in goats during the advance period of West Africa Sahelian dry season. *Wld. Rev. Anim. Prod*. 19: 49-54.

[3] Aliyu, J. (2007). Assessment of the productivity in the four strains of indigenous chickens in a semi-arid region of North-eastern Nigeria. Unpublished Ph.D. Thesis, proposal. Department of Animal Science, University of Maiduguri, Maiduguri.

[4] Archetti, I., Tittarelli, C., Ceioli, M., Brivio, R., Grilli, G. and Lavazza, A. (2008).Serum chemistry and haematology values in commercial rabbits: Preliminary data from industrial farms in Northern Italy. 9th*World Rabbit Congress*. June 10-13. Verona. Italy.

[5] Asuquo, B.O. (1997). Nigeria journal of Animal production24 (**1**): 46-50

[6] Bush, B.M. (1975). *Veterinary laboratory Manual*. William Heinemann Medical Books Ltd. London. 442p.

[7] Canadian Council on Animal Care (1984). Guide to care and use of pets and laboratory experimental animals. 1:85-86.

[8] Duncan, D.B. (1955). Multiple range and multiple F-tests. *Biometrics* 11: 1- 42.

[9] Ehebha, T.E., Omoikhoje, S.O., Bamgbose, A.M., Aruna, M.B. and Isidhahomen, C.E. (2008). Haematology and serum chemistry of weaner rabbits fed cooked Bambara groundnut meal as replacement for soya bean meal. *Proc. 33rd Ann. conf, Nig. Soc. Anim. Prod*. (NSAP). Pp. 192-194.

[10] Encarta. (2007). Encarta kids. Encyclopedia by Microsoft incorporations. USA.

[11] Medirabbit.com (2003). Complete blood count and biochemistry reference values in rabbits. Accessed online at www.medirabbit.com on 19/2/2011.

[12] Omoikhoje, S.O., Bamgbose, A.M., Oboh, S.O. and Eghiaruwa, B. (2004). Replacement value of groundnut cake with Bambara groundnut on the haematological traits and serum biochemistry of broilers. *Proc. 9th Ann. Conf. Anim. Sci. Ass. Nig*. (ASAN).13th-16th Sept. 2006. Abakaliki, Nigeria Pp. 86-89.

[13] Omoikhoje, S.O., Bamgbose, A.M., Aruna, M.B. and Uwagbale, A.A. (2005). Response of starter broiler fed graded levels of cooked Bambara groundnut meal. *Proc.39thconf. Agric. Soc. Nig.*, University of Bennin, Benin Pp. 120-121

[14] Omoikhoje, S.O., Bamgbose, A.M.and Aruna, M.B. (2006). Growth response and cost benefit of broiler finisher fed diets containing different levels of cooked Bambara groundnut meal. *Proc.11thAnn. Conf. Anim .Sci. Ass. Nig*. (ASAN).Sept.18th-21st.I.A.R & T. Ibadan, Nigeria. Pp. 158-159.

[15] Schalm, O.W., Jain, N.C. and Carrol, E. (1975). *Veterinary Haematology* 3rd ed. Lea and Febiger Publishers, Philadelphia, USA. Pp. 160-210.

[16] Steel, R.G.D. and Torrie, J.H. (1980).*Principles and Procedures of Statistics:A biometrical approach.* 2nd ed. McGraw Hill Book Co. New York, USA.

[17] Ugherughe, P.O. and Ekedolum, P.A. (1986). Pasture and rangeland potentials of Borno. *Annals of Borno* 3: 179-192.

[18] WHO. (1980). *Manual of Basic Techniques for a Health Laboratory*. World Health Organization, Geneva, Switzerland Pp. 75-43

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

Optimizing the Irrigation Water Needs of Lebak Semendawai Swamp in Increasing Agricultural Production

Dinar DA Putranto[1*], Sarino[2], Agus Yuono[3], Agus Karsa Yudha[4]

[1,2,3]Department of Civil Engineering and Planning, Faculty of Engineering, University of Sriwijaya, Indonesia
*Email: dinar.dputranto@gmail.com
[4] Department of Public Works, OKU Regency, South Sumatra, Indonesia

Abstract— *East OKU Regency, South Sumatra Province, Indonesia, is a region that has great potential in the agriculture and plantation sectors. Utilization of swamps and tidal swamps is used as an alternative to increasing agricultural yields despite extreme changes in river water flow downstream during the dry season.*
This study aims to analyze the magnitude of the potential discharge mainstay, and the influence of the magnitude of the flow of the Komering river flow to the availability of water and the availability of optimum discharge in the Lebak Semendawai irrigation area. The total area of 1,218.83 hectares of rice fields, 374.9 hectares is a shallow swamp.
Based on rainfall data for the last ten years, it shows that the potential for discharge is 2.67 m3 /sec, while the required water needs is 2.16 m3/sec. (excess water is 0.51 m3/sec). The results of the analysis show that the planned cropping patterns that can be applied are Paddy - Paddy – Secondary Crop. The Komering river water discharge which affected the first cropping rice planting pattern was 62.877 m3/sec, the second rice planting period was 43.41 m3/sec and during the cropping period the water demand could be fulfilled, if it was achieved through pump system with a capacity 1,657.6 liters/sec because the water level of the river from June to November are under the baseline elevation of floodgate on retrieval buildings. Water requirements for the entire irrigation network system in the Lebak Semendawai marsh swamp are 37.22 m3/sec.
Keywords— ***Agricultural, discharge, irrigation, swamp, water level.***

I. INTRODUCTION

The Indonesian Government's efforts to increase rice productivity include building a swamp irrigation network. Irrigation area of Lebak Semendawai is a rainfed rice field developed by the Indonesian government in South Sumatra Province with an area of approximately 2,244 ha. But in its development, in one year, farmers plant rice between one and two times, but in the second planting season the possibility of failure is more dominant because it has entered the dry season, making it difficult to get water. Water sources are located around the irrigation area. The reliable Lebak Semendawai is the Komering River.

This study aims to analyze the magnitude of potential discharge mainstays in the Lebak Semendawai area, and analyze the magnitude of the Komering river flow discharge to the availability of water as a source of Lebak Semendawai irrigation water, as well as analyze the availability of optimum discharge for the Lebak Semendawai irrigation flow.

II. METHODOLOGY

2.1 Research Area

The Lebak Semendawai irrigation area is administratively located in Campang Tiga Ulu village, Sukaraja village and Gunung Jati village, Cempaka District, East OKU Regency, South Sumatra Province with geographical coordinates of east longitude 104º10'1.2" - 104º41'49.2" and 4º27'32.4" - 4º27'32.4" south latitude.

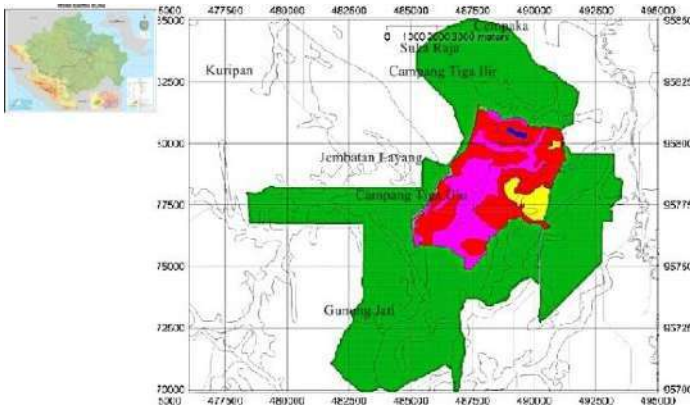


Fig. 1: Lebak Semendawai location area

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

2.2 Research Methods

To analyze the availability and demand for water resources in the study area, several equations are used to perform calculations

2.2.1 Rain plan

Calculation of rain plan is done using several distribution methods, namely the Normal Method, Normal Log, Gumbel, and Pearson Type III log [1]

Normal distribution method,

$$X_T = \mu + K_T.\sigma \quad \text{.....(1)}$$

with

XT : Rain plans with a T year return period;
μ : Average values of rain data (mm);
σ : Standard deviation.

Normal Log Distribution Method,

$$Y_T = \overline{Y} + K_T.S \quad \text{.....(2)}$$

With,

Y_T : Estimated value expected to occur with a T-annual return period;
$\overline{Y}$: The average value of the variate count;
S : Standard deviation of variate values;
K_T : Frequency factor.

Pearson Log Type III distribution method,

$$\overline{\log X} = \log \overline{X} + G*S \quad \text{.....(3)}$$

Gumbel distribution method,

$$X = \overline{X} + \frac{y_T - y_n}{S_n}\sigma_n \quad \text{.....(4)}$$

With,

X : Extreme value;
$\overline{X}$: average value;
y_T : reduced variate;
y_n : reduced variate mean;
σ_n : standard deviation

To find an appropriate design, the Chi-Square Test was conducted to test the suitability of the distribution. Chi Square Test (X^2) was carried out using the following equation,

$$X^2{}_{count\ it} = \sum_{i=1}^{k} \frac{(Fe - Ft)^2}{Ft} \quad \text{.....(5)}$$

$X^2{}_{count\ it}$: price *Chi-Square* count it;
Fe : Frequency of observation j class;
Ft : Frequency of Frekuensi teoritical j class;
k : Class Total.

2.2.2 Calculation of peak discharge

To design the amount of peak discharge in the study area is carried out using the following approaches,

Nakayasu HSS formula for Flood Peak Discharge,

$$Qp = \frac{c.A.R_O}{3{,}6(0{,}3Tp + T_{0,3})} \quad \text{.....(6)}$$

with

Qp : Qmaks, is the peak flood discharge (m3 / sec);
c : flow coefficient (= 1);
A : Watershed area (until to *outlet*) (km²);
Ro : unit rain (mm);
Tp : the grace period from the beginning of the rain to the peak of the flood (hours);

While the Rational method function is used to determine the design flood discharge, namely by the equation[2],

Q = 0,278 C.I.A (A in ha) .....(7)

With,

Q : Design flood discharge (m^3/sec);
C : Flow coefficient;
I : rain intensity (mm/hour);
A : Watershed area (km^2 or ha).

While the flood discharge equation according to Haspers [3]

Q = α. β. q. F .....(8)

with,

f : chatment area (km^2);
α : drainage coefficient;
β : reduction coeficient;
q : maximum rainfall (m^3/km^2/sec).

And the flood discharge equation according to Mononobe [4]

$$Q = \frac{\alpha.r.f}{3{,}6} \quad \text{.....(9)}$$

With,

α : drainage coefficient;
r : rainfall intencity (mm/hour);
f : chatment area (km^2);
Q : Flood discharge (m^3/sec).

While the flood discharge equation according to Melchior [5]

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

$$Q = \alpha.x.I.x.A.x.\frac{r}{200} \quad \text{.....(10)}$$

With,

r : Maximum daily rainfall (mm).

To find out the intensity of rain with a specific period design used the Mononobe method rainfall intensity equation [6]

$$I = R_{24} / 24\left[24/t\right]^{n} \quad \text{.....(11)}$$

I :The intensity of rainfall (mm/hours);

t : rain concentration time (hours), for Indonesia 5~7 hours;

R_{24} : maximum rainfall of 1 day (mm/hours);

n : constants (for Indonesia estimated n~2/3).

Note: the reset factor factor is entered in R_{24}.

2.2.3 Irrigation Water Needs

Estimation of irrigation water requirements is carried out by taking into account the guidelines of the Department of Public Works[7],

(1) The need for clean water in the rice fields

NFR = Etc + P – Re + WL

(2) Irrigation water needs for rice, WRD

IR = NFR / e

(3) Need for land preparation for rice

(4) Irrigation water needs for secondary crops, WRP

IR = (Etc - Re) / e

Where,

Etc : Consumptive Use;

P : Water loss due to percolation (mm/day);

Re : Effective rainfall (mm/day);

E : Overall irrigation efficiency;

WLR : Water layer replacement (mm/day).

The amount of evapotranspiration is used by the Penman modification method [8]

ET = C [w.Rn + (1-w) f(U) (ea – ed).....(12)

With,

ET : Evapotranspiration (mm/day);

C : Correction factors due to climate conditions day/ night;

Rn : Net radiation is equivalent to Evaporation (mm / day);

Director General of Irrigation Department of Public Works[9] states that in general water losses in irrigation networks can be grouped into :

(a) Between 15% to 22.5% in tertiary plots, between tertiary tapping buildings and rice fields;

(b) Between 7.5% to 12.5% in the secondary channel, and;

(c) Between 7.5% to 12.5% in the primary canal.

Calculation of irrigation needs during land preparation can use the method of Van De Goor and Zijlstra[7] (Directorate General of Irrigation Department of Public Works, 1986), namely:

$$IR = \frac{Me^{k}}{(e^{k} - 1)} \quad \text{.....(13)}$$

where

IR : Irrigation water needs at the level of rice fields (mm / day);

M : Water needs to replace water losses due to evaporation and percolation in saturated fields M = Eo + P (mm/day)

Eo : Open water evaporation taken 1.1 Eto during land preparation (mm/day);

P : Percolation;

k = MT/S.....(14)

T : Time period for land preparation (days);

S : Water requirements, for saturation are added with a layer of water 50 mm, ie 200 + 50 = 250 mm.

Consumptive use is calculated using equations [8]

Etc = Kc . Eto.....(15)

with,

Etc : Plant evapotranspiration (mm/day);

Eto : Reference crop evapotranspiration (mm/day);

Kc : Crop coefficient.

The rate of percolation is very dependent on the properties of the soil. In clay soils, with good processing characteristics, percolation rates can reach 1-3 mm/day[10] Effective rain is rainfall that can be effectively utilized by plants. For irrigation in rice plants, monthly effective rainfall is taken 70% of the average monthly rainfall with a possibility of not meeting 20%

$Re = 0,7 \times R_{80}$ .....(16)

With,

Re : Effective rainfall (mm/day);

R_{80} : Mid average monthly rainfall with a 20% chance of not being met.

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

Planting patterns in one year must see the presence or absence of water (water availability) in irrigated areas[11]
Conventional discharge measurements can be done by

(1) Determine the wet cross-sectional area of the river (A), i.e. by measuring the estimated water;
(2) Measuring water velocity (V) with a speed meter (current meter) or buoy (the speed is measured with a Stop watch).

Then the discharge calculation (Q) is performed as follows[12]

$A_1V_1rata^2 + A_2V_2rata^2 +...+ A_nV_nrata^2 = Q$.....(17)

with,

Q : River discharge (m^3/sec);
A_n : N-river cross-sectional area (m^2);
$V_{n\ rerata}$: Average speed on n-cross section (V_{rerata} point 0,2 h and 0,8 h).

Mainstay debits are debits available throughout the year with a certain risk of failure. Mock introduced a simple model of simulation of monthly water balance for flow which includes rainfall data, evaporation and hydrological characteristics of drainage areas[8]

Ea = ETo - ΔE → (Ea = Et) .....(18)
ΔE = Eto x (m/20) x (18–n)→(E=ΔE).....(19)
with :
Ea : Actual evapotranspiration (mm/day);
Et : Unlimited Evapotranspiration (mm/day);
Eto : Potential evaporation of the Penman Method (mm / day);

Basic planning with regard to land units is tertiary plots. These compartments receive irrigation water that is flowed and measured in tertiary off take structures. The secondary plot consists of several tertiary plots, all of which are served by one secondary channel. The primary plot consists of several secondary plots which take water directly from the primary channel.
The discharge plan for a channel is calculated by the formula [11]

$$Q_t = \frac{NFR.A}{1000.e_t} \quad \text{.....(20)}$$

Where,

Q_t : Discharge plan (m^3/sec);
NFR : The need for clean water in the fields (lt/sec/ha);
A : The area of water is irrigated (ha);
e : Irrigation efficiency in tertiary plots.

Overall efficiency (total) is calculated as follows:
Tertiary network efficiency (et) x Secondary network efficiency (es) x Primary network efficiency (ep), and between 0.65 - 0.79 [7]

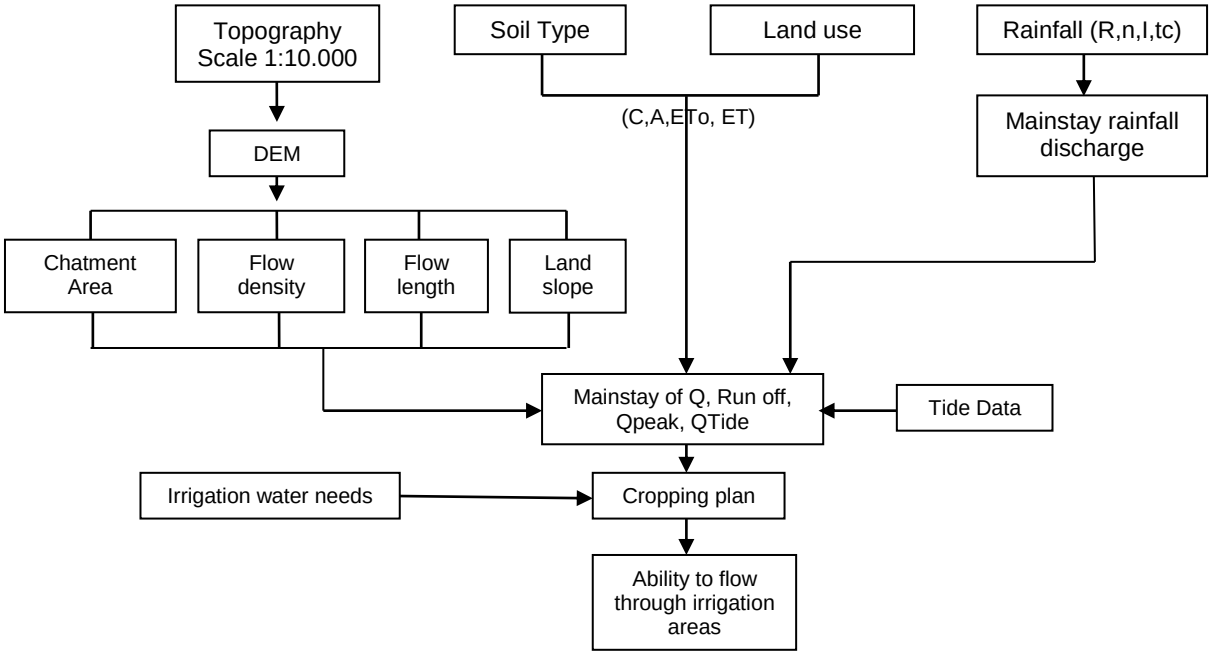


Fig. 2: Research flow chart

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

III. RESULTS AND DISCUSSION

3.1 Data Discharge Input (Inflow)

The initial data sources used in the analysis are rainfall distribution data and Komering river water discharge data. Rainfall data used to forecast rainfall is daily rainfall data for the years 2005 - 2014 obtained from the Seed center Directorate General of Agriculture BK 10 Gumawang Station, East OKU Regency.

The maximum average rainfall between 2005 and 2014 based on Figure 3 below occurred in January of 364 mm while the minimum rainfall between that year occurred in July and August of 81.1 mm.

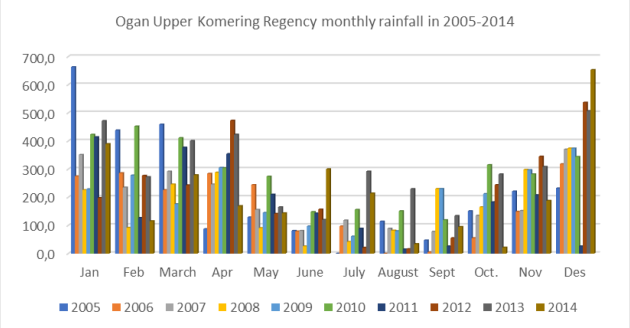


Fig. 3: Monthly maximum rainfall data

3.2. Plan Rain Analysis

The return period that will be calculated in each method is the return period of 2, 5, 10, 25, 50 and 100 years. The rainfall data used is the maximum daily rainfall data.

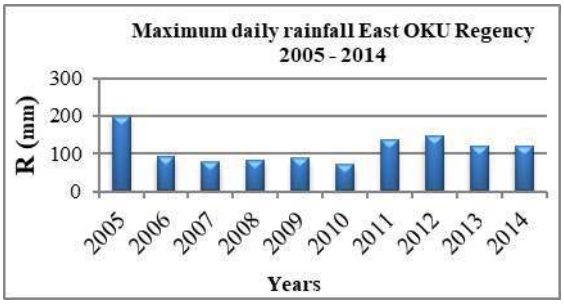


Fig. 4: Maximum daily rainfall data

Figure 4 above provides information that the highest maximum daily rainfall data occurred in 2005 of 199 mm and the lowest occurred in 2010 of 71 mm.

Table 1. Results of calculated rainfall plans

T (Years)	Frequency distribution of rainfall R(mm)			
	Normal	Log Normal	Log Person III	Gumbel
2	113,45	107,89	110,92	108,11
5	146,68	141,91	140,61	155,26
10	164,09	164,06	166,34	186,55
25	181,02	188,79	201,37	226,04
50	194,54	210,86	229,62	255,36
100	205,62	231,21	259,42	284,44

The rainfall plan (R) in the table above can be displayed in the graph below,

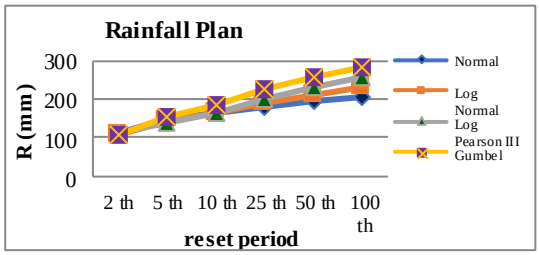


Fig. 5: Rainfall Frequency Distribution

Distribution test results can be seen that the distribution that meets the criteria is the Pearson Log Type III Distribution and Gumbel Distribution.

Table 2. Distribution Test Results

Distribution Type	Terms	Calculation	Conclusion
Normal	Cs=0	Cs=1,125	Not meet
	Ck=3	Ck=4,972	
Log Normal	Cs=3CV+(CV2)=3	Cs=0,212	Not meet
	Ck=5,383	Ck=3,555	
Log Person III	Cs≠0	Cs=5,536	fulfill
Gumbel	Cs≤1,396	Cs=1,125	fulfill
	Ck≤5,4002	Ck=4,972	

3.3. Chi Square Test

Chi Square Test Results of Pearson Type III Log distribution and Gumbel distribution obtained the parameter Chi-Critical Square (X^2cr) = 7.815 for degrees of freedom (df) = 3 and the level of confidence (α) = 5% so that the price of Chi Square is calculated (X^2 count) = 0,4. Because X^2 counts are smaller than X^2cr, it means that the data corresponds to both distributions. Because the Variance Coefficient (CV) of the Pearson Type III Log distribution is smaller than the Gumbel distribution, the Pearson Type III Log distribution will be used.

3.4. Mainstay Rainfall

The rainfall distribution method that can be used from the distribution test results is the Log Pearson III method. The result is obtained a reliable rainfall of 80% (R80) 183.65 mm. Thus it can be concluded that the 80% reliability opportunity occurs at a probability of 30%, ie between 180 mm and 191.9 mm of rainfall.

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

3.5. Rainfall Intensity

The intensity of rainfall (I) and daily maximum rainfall (R_{24}) of the calculation results can be explained in the graph below,

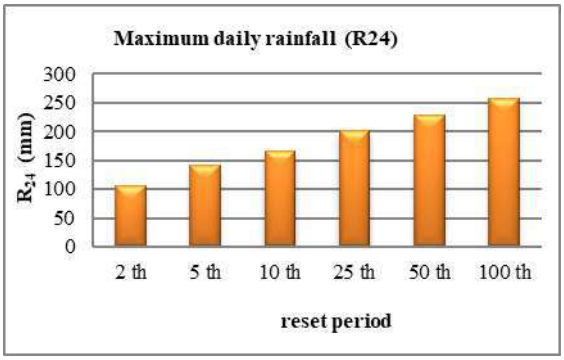


Fig. 6: Daily Maximum Rainfall

The data in Figure 6. above states that for a return period of 2 years the maximum daily rainfall (R_{24}) is 104.71 mm and for a 100 year return period of 258.05 mm. The amount of rainfall intensity (I) that is influenced by daily maximum rainfall (R_{24}) can be seen in the graph below

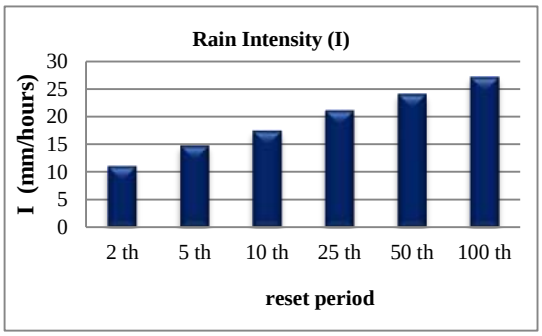


Fig. 7: Intensity of rainfall Mononobe method

The intensity of rainfall (I) that occurred in the 2-year return period is shown in Figure 6 above 10.994 mm / hour and the 100-year return period of 27,093 mm/hour.

3.6. Mainstay Discharge

Analysis of rainfall with the probability method is known that rainfall data and the number of rainy days (HH) that can be used is rainfall data in 2008. From the calculation results obtained by the reliable discharge as follows

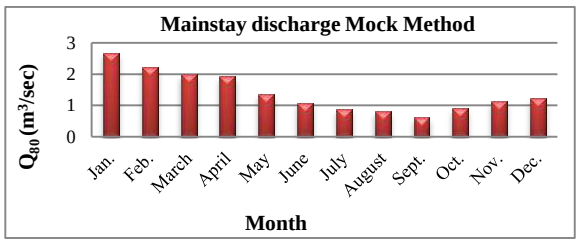


Fig. 8: Mainstay Discharge Method F.J. Mock

The magnitude of the mainstay discharge stated in Figure 8 above is the highest mainstay discharge occurred in January which is 2.67 m^3/sec and the lowest occurred in September of 0.61 m^3/sec. This means that the decline in reliability occurs from January to September.

3.7. Nakayasu Synthetic Hydrograph Unit

The results of the calculation of the Nakayasu Synthetic Hydrograph can be obtained from the magnitude of flood discharges of the return period of 2 years to 100 years.

The Nakayasu method flood discharge at Table 3 can be outlined in the form of a hydrograph model shown in the following Figure 9,

Table 3. Nakayasu Method Flood Peak Discharge

Re Period T (years)	Flood peak discharge (m^3/sec)
2	200,974
5	269,489
10	318,290
25	385,200
50	438,759
100	495,500

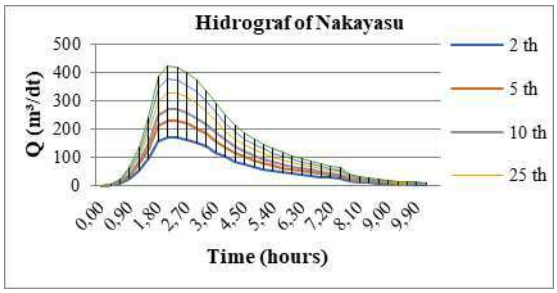


Fig. 9: Nakayasu Synthetic Hydrograph Lebak Semendawai irrigation

3.8. Flood Peak Discharge

Calculation of peak discharge (Qp) with the Rational method obtained through spatial analysis can be displayed in the figure below

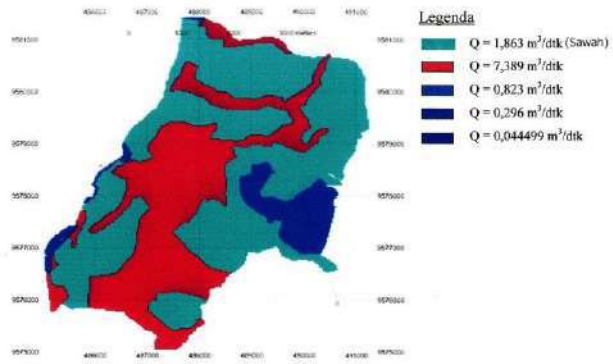


Fig. 10: Peak Discharge (Qp) Rational method

Data processing from the spatial analysis results above shows that the highest peak discharge (Qp) at the location

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

of the study was 7.389 m^3/s and the lowest was 0.044499 m^3/s. The total peak discharge (Qp tot) that occurred was 10,415 m3/sec.

3.9. Peak Flood Discharge Plan with the Empirical Method

The calculation of flood discharge plan is calculated by empirical methods including Haspers, Mononobe and Melchior methods, the following results are obtained.

Table 4. Recapitulation of Flood Hydrograph Calculation Results for Empirical Method Planning

Period	flood discharge (Q) (m^3/sec)		
T(years)	Haspers	Mononobe	Melchior
2	97,066	64,187	71,417
5	130,040	85,992	95,678
10	153,669	101,617	113,063
25	185,947	122,962	136,812
50	211,829	140,077	155,855
100	239,212	158,185	176,003

Flood discharge (Q) Empirical method in Table 4. above can be poured into the following graph

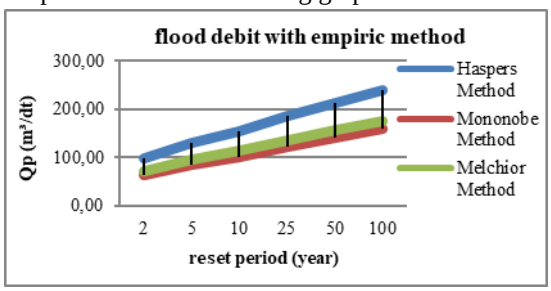


Fig. 11: Flood Hydrograph of the Empirical Method Plan

3.10 Irrigation Water Needs Analysis

3.10.1 Evapotranspiration of the Penman Method

Calculation of potential Evapotranspiration (ETo) with the Penman method obtained the following results

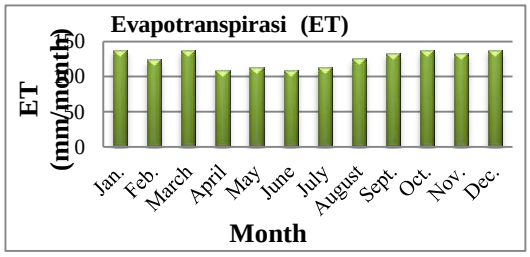


Fig. 12: Evapotranspiration of the Modified Penman Method

Figure 12 above explains that the highest Evapotranspiration occurred in January, March, October and December amounted to 137.90 mm/month and the lowest occurred in April and June amounted to 109.19 mm/month. The amount of Evapotranspiration (ET) based on land use can be shown in the following graph.

The highest evapotranspiration based on Figure 13. The follow occurred in the cocoa area which was 1.42 mm/day and the lowest occurred in the rice field area 0.20 mm/day. This shows that evapotranspiration in paddy fields is lower than other areas

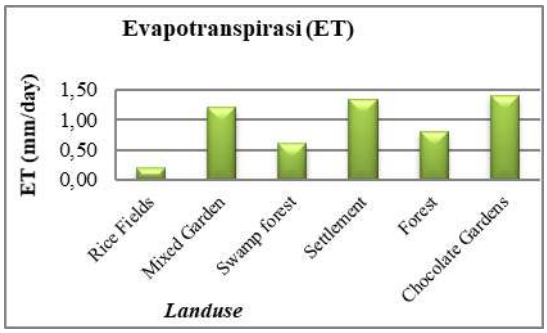


Fig. 13: Evapotranspiration based on Land Use Maps

3.10.2 Percolation

The rate of percolation is very dependent on the properties of the soil. The percolation rate used is 2 mm/day with consideration of soil texture in the location area

3.10.3 Effective rainfall for rice and secondary crop water needs

The probability in determining how much the reliability of the flow is applied using the basic year method. The results of these methods obtained by reliable rainfall of rice plants (R_{80}) using rainfall data in 2011 and reliable crops of crops (R_{50}) using rainfall data in 2014.

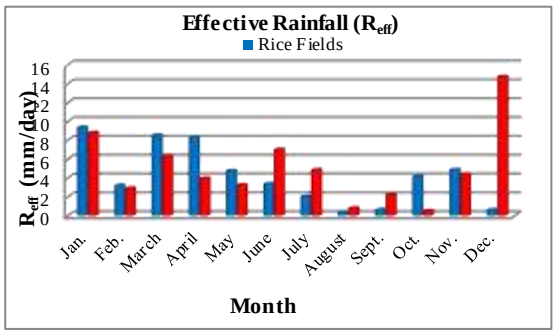


Fig. 14: Effective rainfall of rice and secondary crops

Maximum effective rainfall (Reff) for rice analysis results Figure 14 above is estimated to occur in January which is 9.35 mm day and the maximum reff for secondary crop occurs in December amounted to 14.74 mm/day.

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

3.10.4 Irrigation Efficiency

The guideline used for irrigation efficiency planning in operation and implementation, namely water loss in Tertiary plots is determined 20% between tertiary tapping buildings and paddy fields with an efficiency factor of 1.25. In the Secondary Channels is determined 10% with an efficiency factor of 1.11 and for the Primary Channels is determined 10% with an efficiency factor of 1.11.

3.10.5 Irrigation Water Needs and Planting Patterns

Irrigation water needs include crop water needs per irrigated land area. Planting patterns that can be applied from the analysis of irrigation water needs are Rice - Rice – Secondary Crop with the provisions that the Rice planting period lasts for 4.5 months, Rice planting period for 4 months and crops for 3.5 months. The results of the analysis of the calculation of Irrigation Water Needs for an area of 1,218.83 Ha can be displayed in the graph below

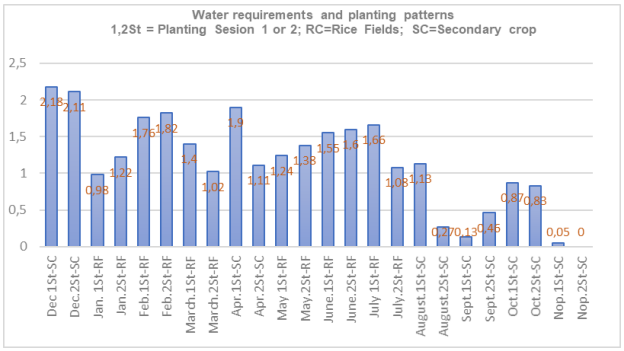


Fig.15: Irrigation Water Needs and Planting Pattern Plans

The amount of maximum irrigation water requirements for the 1st planting season for 1,218.83 Ha as shown in Figure 15 above is 2.18 lt/sec/ha at the beginning of December, the 2nd planting period 1.90 lt /sec/ha in early April and secondary crop at 1.13 lt/sec/ha in early August

3.11 River Water Discharge Based on Comparison of Rainfall Occurred

River water discharge (Q) when rainfall is low in October 2014 results of measurements of river flow velocity using Current meters and river cross section using Echosounder is 25.30 m^3/sec. River water discharge (Q) during moderate rainfall of 219.97 m^3/sec is obtained based on the ratio of river water discharge and rainfall that occurs when rainfall is low in October with moderate rainfall in November.

The results of comparison of river water discharge and low rainfall in October with the prediction of high rainfall in January obtained river water flow (Q) when high rainfall amounted to 426.35 m^3/sec.

3.12 Potential Use of the Komering River as a source of Lebak Semendawai Irrigation Water

3.12.1. Water Levels Based On Water Level Fluctuations

The results of data processing of water level fluctuations show that the average water level is 1.156 m, the highest high tide is 1.65 m and the lowest low tide is 0.75 m. Fluctuations that occur are mixed tides (0.25 <F <3.00) based on the Formzahl value F = 1.18 above. Of the two types of mixed tides, the tides that occur are the mixed-dominant diurnal tides for 0.50 <F ≤ 3.00.

3.12.2 River water discharge based on prediction of high fluctuations in water

River water discharge is obtained from the multiplication between the total river cross-sectional area (A) and the river flow velocity (V). The measurement results using Echosounder and Current Meter as well as water level from river water level fluctuation data can be seen that,

(1) Based on the average height of low water level over a period of 19 years (MLWL) 0.59 m, obtained river water flow (Q) when rainfall is low at 24.37 m3/sec;
(2) Based on the average height of the high water level over a period of 19 years (MHWL) 1.57 m, the river water flow obtained during moderate rainfall was 144.7 m3/sec;
(3) Based on the highest water level at the tidal full moon (HHWL) 1.86 m, river water discharge obtained during high rainfall amounted to 209.59 m3/sec;

3.12.3 Prediction of availability of average river water discharge per month based on fluctuations in surface water and rainfall occurred

The average river water discharge (Qrt) per month can be displayed in the following graph

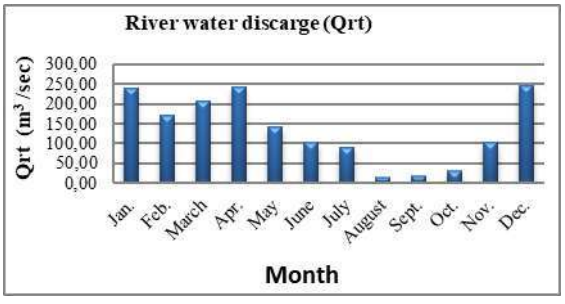


Fig. 16: Monthly river water discharge (Qrt)

The highest average river water flow (Qrt) based on Figure 16 above occurred in December, which was 246.60 m3/s and the lowest occurred in August 14.36 m^3/s. The predicted results of the average water level per month can be displayed in the following table

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

Table 5. Monthly Average River Water Level

Month	Average Water Level (m)
Jan.	2,60
Feb.	1,84
Marc.	2,23
Apr.	2,65
May	1,53
June	1,11
July	0,98
Aug.	0,16
Sept.	0,20
Oct.	0,34
Nov.	1,11
Dec.	2,67

The water level above when connected to the condition of the building intake (free intake) can be shown in the following figure

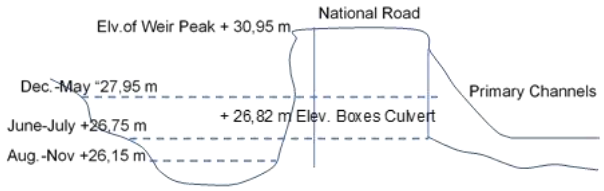


Fig. 17: Average River Water Level against Free Intake Conditions

River water discharge in June - July and August - November based on Figure 16 above has an average water level of 1.05 m and 0.45 m from the bottom of the river under the elevation of the Box Culvert base so as to be able to drain the irrigation water source to the location Paddy fields cannot use box culvert channels to drain water, so a pump system is needed.

3.13 Water Discharge Irrigation Network System

How much water flow (Q) needed to irrigate the irrigation network must be analyzed

3.13.1 Water availability per month based on planned water needs and cropping patterns

How much water flow (Q) needed to irrigate the irrigation network must be analyzed how big is the availability of water available, compared with the required water debit. The results of the calculation of water discharge (Q) needed based on the analysis that has been done is 37.220 m3/sec. to flow through 1271.69 Ha of paddy fields.

(a) Paddy planting season 1 (end of November - March when rainfall is high)
River water discharge during high rainfall 209.59 m³ /sec; Availability of river water at the intake for irrigation 103.57 m3/sec; Peak water discharge on the surface 10,415 m3/sec; availability of water for irrigation (30% x 209.59) + (70% x 10,415) = 70.17 m3 /sec;

(b) 2nd planting season (April - July during moderate rainfall)
River water discharge during moderate rainfall 144.70 m³/sec; Availability of river water at the intake for irrigation 38.68 m3/sec; Water discharge rivers for irrigation in June and July (- 3.48 m3/sec and - 15.3 m3/sec) are met if pumping with a capacity of 1657.6 lt/sec; Peak water discharge on the surface 5,705 m3/sec; availability of water for irrigation (30% x 144.70) + (70% x 5.705) = 47.40 m3 /sec;

(c) Secondary crop (August - early November when rainfall is low)
River water discharge when rainfall is low 24.37 m³ / sec; Availability of river water for irrigation does not exist - 81.65 m3/sec (Qintake = 0.00 m3/sec); River water discharge for irrigation is fulfilled if the pump capacity is 1,657.6 lt/sec; River water discharge for pumping irrigation resulting from 7,311 m3/sec; Peak water discharge at 4,517 m3/sec; Water supply for irrigation is 11,828 m3/sec (7,311 m3/sec + 4,517 m3/sec).

3.13.2 Excess water in the swamp area and adjustment of the planting pattern plan

The plan to prepare land in swampy swamp area for planting rice 1 is strongly influenced by excess water in the area because it is not possible to plant if the amount of water is too excessive. The excess water for the 1st planting season rice in the swampy swamp areas above based on the analysis of water availability in the plot can be shown in the following table

Table 6. Excess water in the plot and water discharge plots in the swamp area based on water availability for the 1st Planting Rice

Rice Fields	Excess water plot plan (m3/sec)	Swamp water discharge (m3/sec)
CT4-Ka	0,268	0,232
CT4-Tg	0,177	0,286
C1-Ki	0,180	0,169
C2-Ki	0,191	0,239
C3-Ki	0,194	0,282
C3-Ka	0,199	0,235
Lb.4-Ki	0,136	0,194
Lb.4-Ka	0,264	0,293
Lb.3-Ka	0,180	0,225
Lb.3-Ki	0,077	0,071
Total	1,866	2,226

https://dx.doi.org/10.22161/ijeab.51.17 *ISSN: 2456-1878*

The excess water in the planned plot of swampy swamp area according to Table 6 above is 1.866 m3 / sec (161.222.4 m3 / day). Water discharge in the swampy swamp area is 2,226 m3 / sec (192,326.4 m3 / day) with a swampy swamp area of 374.9 Ha of the total planned plot of 663.05 ha. The water discharge is predicted to occur at the highest rainfall in December. Thus, monthly water discharge in the swampy swamp area based on comparison of the average rainfall that occurs can be displayed in the following table

Table 7. Prediction of water discharge in the swamp area per month

Month	Water discharge in swampy area (m3/det)
Jan.	2,169
Feb.	1,534
Marc.	1,854
Apr.	1,748
May	1,013
June	0,734
July	0,649
Aug.	0,483
Sept.	0,607
Oct.	1,051
Nov.	1,459
Dec.	2,226

The highest prediction of swamp water discharge based on Table 7 above occurred in December, which was 2.226 m3/sec (192,326.4 m3/day) and the lowest in August was 0.483 m3/sec (41,731.2 m3/day). Planting patterns that can be applied to the swampy swamp area are based on the above water discharge, namely:

(1) The first cropping season in October - February;
(2) The 2nd cropping season in March - early June;
(3) Secondary crop at the end of June - September.

The cropping plan for the 1st planting season is planted with special local species, namely surung rice (alabio, tapus, nagara and hiyang) because the water discharge is still quite high. For the second planting season rice can be planted with rice types in Indonesia (IR 42, IR 64, IR 66, and local species as Cisokan, Ciherang, Cisanggarung and Mekonga rice types). Secondary crop can be planted with a mound system (elevated section). Rice fields in the form of rice fields in general (not swamp) can follow the previous cropping plan, namely:

(1) The first cropping season at the end of November - March;
(2) Rice planting period 2 in April - July;
(3) Secondary crop in August - early November.

IV. CONCLUSIONS AND RECOMMENDATIONS

4.1 Conclusions

(1) Potential magnitude of mainstay discharge based on the swamb area based on the flowing pattern limit in the Lebak Semendawai area which is affected by the Komering River is 2.67 m³/sec with the required water demand of 2.16 m³/sec so that there is excess water (water surplus) of 0,51 m³/sec (sufficient);
(2) The magnitude of the influence of river water discharge is able to support the availability of water for irrigation activities based on the planned Rice - Rice – secondary crop cropping pattern, namely:
   (a) The 1st planting season (end of November - March) takes place during high rainfall, river water discharge (30%) which can be used for irrigation of 62,877 m3 / sec;
   (b) The 2nd planting season (April - July) takes place during moderate rainfall, river water discharge (30%) which can be used for irrigation of 43.41 m3/sec with the irrigation network water requirement of 32.62 m3/sec (sufficient). Irrigation network water needs in June and July can be fulfilled if a pumping system with a pump capacity of 1657.6 lt/sec is implemented, given the river water discharge is below the elevation of the floodgate;
   (c) Secondary crop (August - Early November) takes place when rainfall is low, river water discharge that can be used for irrigation is absent (- 81.65 m3/sec below the elevation of the floodgate at the intake). Surface water discharge is 4,517 m3/s. Irrigation system water needs can be met if pumping is carried out with a pump capacity of 1,657.6 lt/sec so that 30% of river water debit obtained is 7,311 m3/sec. Thus the crop water needs are fulfilled (7,311 m3/sec> 3.74 m3/sec).
(3) The amount of optimum availability of Lebak Semendawai Irrigation is:
   (a) Paddy planting period 1 availability of optimum discharge is 70.17 m3/sec. The availability of water in the plot of rice for 6,472 m3/sec with plant water needs 2,167 m3/sec;
   (b) In the 2nd planting period, the optimum discharge availability is 47.40 m3/sec. The availability of water in the planned plot of rice is 5,027 m3/sec with plant water needs 1,897 m3/sec;
   (c) the availability of optimum discharge Secondary crop 11,828 m3/sec. Availability of water in

planned plots for secondary crop 1,817 m3/sec with plant water needs 1,128 m3/sec.

4.2 Recommendations

Further research is needed regarding the efficiency and economic value of the use of pumps to irrigate the irrigation network system

ACKNOWLEDGEMENTS

Acknowledgments were conveyed to the Research and Community Service Institute of the University of Sriwijaya University, which provided research funding for Higher Education Competitive Grants based on Agreement / Contract 0149.032 / UN9 / SB3.LP2M.PT / 2019 with the chairman of LPPM, on 27 June 2019

REFERENCES

[1] Oregon State University, "Analysis Techniques : Flood Analysis Example with Daily Data (Log-Perason Type III Distribution)," *Streamflow Eval. watershed Restor. Plan. Des.*, pp. 2002–2005, 2005.

[2] S. Stanchi *et al.*, "Soil erosion in an avalanche release site (Valle d'Aosta: Italy): Towards a winter factor for RUSLE in the Alps," *Nat. Hazards Earth Syst. Sci.*, 2014.

[3] A. Jordán *et al.*, "Wettability of ash conditions splash erosion and runoff rates in the post-fire," *Sci. Total Environ.*, vol. 572, 2016.

[4] D. D. Prayuda, "Temporal and spatial analysis of extreme rainfall on the slope area of Mt. Merapi," *Civ. Eng. Forum*, vol. XXI, no. September, pp. 1285–1290, 2012.

[5] C. A. Bana e Costa, P. Antão da Silva, and F. Nunes Correia, *Multicriteria evaluation of flood control measures: The case of Ribeira do Livramento*, vol. 18, no. 3. 2004.

[6] J. Liu, W. Liu, and K. Zhu, "Throughfall kinetic energy and its spatial characteristics under rubber-based agroforestry systems," *Catena*, vol. 161, 2018.

[7] Department of Public Works, > *SPECIFICATIONS For : I STANDARD*, no. September. 1986.

[8] A. Amazirh *et al.*, "Modified Penman–Monteith equation for monitoring evapotranspiration of wheat crop: Relationship between the surface resistance and remotely sensed stress index," *Biosyst. Eng.*, vol. 164, no. September, pp. 68–84, 2017.

[9] P. Panagos *et al.*, "The new assessment of soil loss by water erosion in Europe," *Environ. Sci. Policy*, vol. 54, 2015.

[10] C. D. Cheng, S. J. Cheng, J. C. Wen, and J. H. Lee, "Time and flow characteristics of component hydrographs related to rainfall-streamflow observations," *J. Hydrol. Eng.*, vol. 18, no. 6, pp. 675–688, 2013.

[11] S. Ali *et al.*, "Planting patterns and deficit irrigation strategies to improve wheat production and water use efficiency under simulated rainfall conditions," *Front. Plant Sci.*, vol. 8, no. August, 2017.

[12] S. Sulianto, M. Bisri, L. Limantara, and D. Sisinggih, "Performance of The Dispin Models with Automatic Parameter Calibration on The Transformation of Rainfall to Runoff Data," *Civ. Environ. Sci.*, vol. 002, no. 02, pp. 084–094, 2019.

https://dx.doi.org/10.22161/ijeab.51.18 *ISSN: 2456-1878*

The Economics of Processing Cashew Products in Benue State, Nigeria

Ogah, Odey Moses; Ogebe, Francis Ozoko and Ukpur, Sandra

Department of Agricultural Economics, Federal University of Agriculture, Makurdi, Nigeria
Corresponding Author: moses4ogah@gmail.com

Abstract**—** *This study was on Economic of Cashew nut processing in Benue State, Nigeria. Simple random sampling technique was used to select one-hundred and twenty-five Respondents. Primary data were used, and collected using a structured questionnaire. Descriptive statistics such as frequency counts, percentages and mean scores were used; Gross margin analysis was used to analyze cost and returns while linear regression was used to analyse factors influencing cashew processing in the study area. Findings revealed that the mean age of the processors was about 33years, most (60.8%) were females, 49.6% were married and nearly all the respondents had formal education. The mean household size was 5 persons. Mean processing experience was 8 years. Research revealed that cashew processing is profitable in the study area. The cost of labour, cost of purchasing and cost of firewood were found to significantly influenced cashew processing in the study area. It was therefore recommended that government agencies and non-governmental organization should provide inputs resources needed by processors, processors should formed cooperative to access credit inputs, male folks were advised to participate in cashew nut processing as it profitable and processors should seek assistance from the government so as to enable them purchase processing machine.*

Keywords**— Analysis, Cashew, Economic and Processing.**

I. INTRODUCTION

The cashew tree native of Brazil was introduced to Nigeria between 15th and 16th Century by the Portuguese explorers as noted by [1] cited by [2]. During the past decades, the production of cashew nuts in Nigeria has increased almost six-fold from 30,000 tonnes in 1990 to 176,000 in 2000 [3]. Prior to this, production was relatively static at 25000 tonnes over a period of 25 years from 1965. Nigeria of recent has recognized the potential economic value of cashew and has made a concerted effort to improve production of the crop.

Cashew known botanically as *Anacardium occidentale L.*, is one of the commodities that has given Nigeria recognition worldwide. The average yield of nuts from a mature tree ranges from 7 – 11kg per annum. The tree is capable of living for between fifty and sixty years and produce nuts for about fifteen to twenty years[2].

Nigeria is the 4th largest producer and produces 220,000 tonnes of the 2.1 million tones world production in 2017. Nigeria farmers earned $404 million from the export of the cash crop in 2017 and between 2015 and 2017; they earned $813.05 in foreign exchange from the exportation of cashew [4].

Cashew processing is a very competitive but also a potentially lucrative activity that can and should be exploited by more processors [3]. Processing of cashew nut as noted by them ensure that the kernels are of high value luxury commodity.

The major objective of processing is to remove the valuable cashew kernel from the shell with as little damage as possible as whole kernel command a higher price than the broken pieces.

Cashew processing is a series of unit operations essential to make available, the edible nut. Variations in processing methodology between different manufacturers are attributed to differences in cashew, availability of equipment-type, human resource and fuel source. In Nigeria, most cashew processing units are at rural level. After 1960, unit operations such as roasting, shell liquid extraction and shelling have been mechanized. However, most other processing steps remain as tedious as manual operations [5].

The various processing operations of Cashew are seemed to be performed manually by experienced semi-skilled workers. This is still the case in Benue state, which is one of the producing states in Nigeria. Since the 1960s, various

https://dx.doi.org/10.22161/ijeab.51.18 *ISSN: 2456-1878*

mechanized cashew pieces of equipment have been developed and are available in several countries [6]. The processes that have been mechanized are roasting, cashewnut shell liquid extraction and shelling. For the most part, the cleaning of raw materials and sizing and kernel grading have remained labour intensive manual operations. There are significant differences in investment requirements, labour skills, health requirements and levels of efficiency between the manual technology and the medium to large-scale mechanical and semi mechanical operations [6]. In general the processing system involves lower investment and variable costs and achieves far greater efficiency in terms of kernel materials yield and the proportion of whole kernels extracted. However this system requires large numbers of experienced workers who work at unhealthy levels of exposure to some hazard [6]. The mechanized systems are more vulnerable to breakdown due to shortage of spare parts require large volumes of nuts for efficient operation and operate well below manufacturer specifications when strict grading and sizing activities are not in place prior to shelling [7] in [6].

Cashew nut processing allows for the development of an important by-product, which can increase its added value. The liquid inside the shell represents 15 percent of the gross weight and has some attractive possible medicinal and industrial uses. CNSL is one of the few natural resins that is highly heat resistant and is used in braking systems and in paint manufacture [6]. It contains a compound known as *anacardium* which is used to treat dermatological disorders. The main markets for cashew nut shell liquid (CNSL) are the United States, the European Union (mainly the Kingdom), Japan and the Republic of Korea. Together these account for over ninety percent of world trade, most of which is supplied by India and Brazil. The cashew nut fruit consists of a peduncle and a seed. The peduncle, often called the false fruit, is pear shaped yellow or red in colour and made up of a soft juicy pulp.The seed which develops below the peduncle is kidney shaped and resembles a large bean. Internally, the seed contains the kernel or cashew nut of commerce surrounded by an oily liquid called cashew nut shell liquid. (CNSL)which is not a triglyceride and contains a high proportion of phenol compounds find its use in industry as a raw material for brake lining compounds as waterproofing agent, a preservative and in the manufacturing of pant and plastics [6]. The kernel contains 47% oil. The main market of cashew nut is as a high value edible nut. The cashew nut shell liquid Cashew apples can also be made into drinks, wines and pickles. Due to the high value of cashew nuts even small pieces find a market in confectionery products [8] in [6].

A lot of studies have been carried out on cashew's production and marketing. For instance, [2] worked on analysis of cashew nut marketing in Kwara state, Nigeria; [9] worked on information delivery and its effects on cashew production in Oyo state, Nigeria; [6] worked on Economic analysis of cashew nut marketing among produce buyers in Ogbomoso metropolis of Oyo state, Nigeria. However to the best of the researcher knowledge, there is little or no work on Economics of processing cashew products, this is the gap the study is intended to fill.

II. MATERIALS AND METHODS

The study was conducted in Gwer local government area of Benue state, Nigeria. Gwer local government which derives its name from Gwer River was created in 1976 out of Makurdi local government with its headquarters at Aliade. It is bordered by Makurdi in the North-East, Gboko in the East, Konshisha in the Southeast, Obi and Oju in the South, Otukpo in the Southwest; and Gwer West in the West. The local government has 14 council wards. Naturally, Gwer local government is endowed with mineral resources which can be effectively tapped by investors.

A multistage sampling technique was used in the selection of the respondents.

Firstly, five (5) council wards namely, Akpach'ayi, Mbaikyaan, Ikyonov, Gbemacha and Mbabur were purposefully selected from the study area based on Cashew processing activities; the second stageinvolved the selection of one community each from the five council wards by means of simple random sampling. The third stage involved the selection of 25 respondents each from the five communities to make total of 125 respondents for the study.

Data for the study were collected from primary source. The data were generated through the use of well-structured questionnaire designed to illicit information from the respondents.

The data obtained were analyzed using descriptive statistics like frequency distribution, percentage, mean and inferential statistics like multiple regression analysis, also gross margin analysis to determine the profitability of the enterprise.

Model Specification.

Implicitly, the model is as specified below

Y= f(CP, CL, CF, CFP, U)

In explicit form

$Y = \beta_0+\beta_1x_1+\beta_2x_2+\beta_3x_3+\beta_4x_4+U$

https://dx.doi.org/10.22161/ijeab.51.18 *ISSN: 2456-1878*

Where Y= Quantity of Cashew processed (Kg)
X_1=Cost of purchasing cashew in naira (CP)
X_2= Cost of labour for processing activities in naira (CL)
X_3= Cost of fire wood in processing cashewin naira (CF)
X_4= Cost of hiring frying pan in naira (CFP)
U= Error term
β_0= constant
βs= Coefficients.
It is expected that the explanatory variables will have inverse relation with the processing of cashew.

III. RESULTS AND DISCUSSION

Socio-Economic Characteristics of the Respondents in the Study Area.

Results of socio-economic characteristic of the respondents are presented in Table1. The result shows that 60.8% of the respondents in the study area were female while 39.2% were male. This implies that more females are involved in cashew processing. This result disagrees with that of [2] findings, who found that 81% of cashew nut processors and marketers were male.[10] noted female's dominance in cashew production in Tanzania. The result of age revealed that the mean age of the respondents was 33.42 years implying that processors are in their productive and youthful age and this could lead to increase in processing of cashew nut in the study area. Specifically, 56.0% of the respondents were between 18-30 years; 24.8% of the respondents were between 31-40 years; 4.0% of the respondents were between 41-50 years, 3.2% were between 51-60 years, while 12.0% were within 61-65 years. This result agrees with that of[11] who reported mean age of 31 for cashew nut farmers in Enugu North, Nigeria. The result revealed that 49.6% of the respondents were married, 46.4% of the respondents were single and 1.6% was divorced, while 2.0% were widowed. This result is in line with that of [2] and[10] this implies that majority of the respondents were married, this is expected because married people are supposed to provide daily meal to their children.

Furthermore, the result show, that 64.0% of the respondent's attained primary education, 22.4% of the respondent's attained secondary education, while 13.6% of the respondents have tertiary education. This implies that, the respondents are knowledgeable and will be open to adopt new technology and innovations. This is in consonance with the findings of [11]. They reported high literacy level for cashew nut farmers.[12] Have noted that the educational profile of the farmer decides the relative exposure of the farmer to latest technologies. The result on years of experience as shown in table1 reveals that the mean processing experience was 8 years which shows that processors had considerable years of experience which is an advantage towards production and adoption of technologies. Specifically, 56.0% of the respondents had 1-5 years of processing experience, 24.8% of the respondents had 11-25 of processing experience. This agrees with[12] and[11] who reported that more experienced processors are knowledgeable and more likely to adopt new techniques.

The result on household size revealed that respondent had a mean household size of 6 persons; specifically 66.4% have 1-5 household size, 27.2% have household size of 6-10, 2.4% have household size of 11-15, while 4.0% of the respondents have household size of 16-25. The result is in line with [2] findings, who reported that majority of cashew nut processors and marketers in Kwara State has relatively large household size. This result implies that majority of the households had large number of members which is an indication of availability of labour for cashew processing. The results on major occupation revealed that majority of the respondents (78.4%) have cashew processing as their major occupation while 21.6% have other occupation other than cashew processing. The result on ownership of cashew farm indicate that majority of the respondent (93.9%) have cashew farm; while 44.8% do buy cashew nut from other source with about 52.6% ownership from inheritance, 5.6% purchased, 6.0% from husband's farm, 4.0% from leasing and 36.6% from other means, also about 8.0% of the respondents that buy cashew nut from outside do buy from agricultural developments programme, 8.0% from market, 84.0% from farmers.

The result also, revealed that about 91.2% of the respondent do hired machine for processing, while about 8.8% have machines of their own.

The result on sources of cashew nut revealed that majority (84%) of the respondent sourced their nut from farmers, 8% of the respondents got their nut from markets, while 8% of them sourced the nuts from Agricultural Development Projects (ADP). The analysis on ownership of processing machine shows that 91.2% of the respondents hired machines for processing activities, while only 8.8% of the respondents have processing machine of their own.

https://dx.doi.org/10.22161/ijeab.51.18 *ISSN: 2456-1878*

Table1: Distribution of Respondents according to their Socio-Economic Characteristic in the Study area n=125

Variables	Frequency	Percentage	Mean
Sex			
Male	49	39.2	
Female	76	60.8	
Age (Years)			
18-30	70	56	
31-40	31	24.8	33
41-50	5	4	
51-60	4	3.2	
61 and Above	15	12	
Marital Status			
Single	58	46.4	
Married	62	49.6	
Divorced	2	1.6	
Widowed	3	2	
Processing Experience (Years)			
1-5	75	59	
11-15	31	24.5	8
21-25	5	4	
31-35	14	11.2	
Educational Status			
Primary	80	64	
Secondary	28	22.4	9
Tertiary	17	13.6	
Household size			
1-5	83	66.4	
6-10	34	27.2	6
11-15	3	2.4	
21-25	5	4	
Ownership of Processing Machine			
Owned	11	8.8	
Hired	114	91.2	

Source: Field survey, 2019

Costs and Returns Analysis in Cashew Processing

Total Variable Cost

This consists of cost of purchasing cashew nut, cost of labour, cost of firewood and cost of frying pan. The result shows that in the study area, the average costs of purchasing 20kg of cashew nut was ₦4435=20, the average cost of labour for processing 20kg of cashew nut was ₦3377=68, also the average cost of firewood for processing 20kg was₦606.80, while the average cost of hiring frying pan for 20kg of cashew nut was ₦3812.40. As revealed from the study the total variable costs for processing 20kg of cashew nut was ₦12,182.08

Total Revenue

https://dx.doi.org/10.22161/ijeab.51.18 *ISSN: 2456-1878*

The average total revenue from the processing of 20kg of cashew nuts was ₦95,733.60.00. The results show that the minimum and maximum revenue were ₦600.00 and ₦1000000.00 respectively.

Gross Margin

Table 2; revealed that the gross margin for processing 20kg of cashew nut in the study area was ₦ 83,516.90. The minimum and maximum gross margin in the study was ₦-4603.00 and ₦983,465.00 respectively. The minimum value of -4603.00 implies that some processors were processing at a loss. Since the average total revenue of ₦83516.90 is higher than the total variable cost of ₦12,182.08, cashew processing is profitable in the study area. This is in line with the findings of [13] that cashew nut processing is profitable in India.

Table.2: Costs and Returns in Cashew Processing in the Study Area

Variables	Min	Max	Mean	Std Deviation
Cost of Purchasing (₦)	500.00	15,000.00	4435.20	3679.75
Cost of Labour(₦)	00	20,000.00	3327.68	4005.99
Cost of fire wood (₦)	50.00	5,000.00	606.80	944.04
Cost of hiring frying pan(₦)	00	17,500.00	3812.40	4258.91
Total Variable cost(₦)	1404.00	35,000.00	12,182.08	8299.40
Total Revenue (₦)	600.00	1,000,000.00	95,733.60	199457.00
Gross Margin (₦)	-4603.00	983465.00	83,516.90	195870.00

Source: Field survey, 2019

Factors Influencing Cashew Processing in the Study Area

The result of factors influencing cashew processing enterprise is shown in table 4, the coefficient of multiple (R^2) is 0.507; indicating that 50.7% of the factors influencing cashew processing is explained by cost of purchasing, cost of labour and cost of fire wood. The result shows that F statistic (24.481) is positive and significant at 1% level indicating the goodness of fit for the model and overall significance of variables used in the model. The result shows that cost of purchasing, cost of labour and cost of fire wood were statistically significant and variables that influence cashew nut processing. [2] Have noted that the cost of purchasing and labour influence the marketing of cashew nut.

Specifically, the coefficient of cost of purchasing is positive and significant at 1%. Level implying that purchasing cost and processing are directly related. The more the purchasing cost the more cashew nuts is being processed. This is not unlikely because the processors will prefer to process the nut at a higher cost to gain more profit.

From the results the coefficient of labour is negative and significant at 1% implying that cost of labour and processing are in inverse relation. The more the cost of labour, the less cashew nut processed and vice versa. Probably, as the cost of labour increases, the processors might make use of family labour to reduce the cost.

Results from table 3, shows that cost of firewood and processing have direct relationship and the coefficient is significant at 1% level.

Table.3: Regression analysis of factors influencing cashew processing in the study area.

Variables	Coefficient	Std. Error	T-Stat	Sign. Level
Constant	4553.042	126.40	3.598	0.000***
Cost of Purchasing	1.341	0.231	5.805	0.000***
Cost of Labour	-0.479	0.183	-2.612	0.010***
Cost of Firewood	6.077	0.810	7.502	0.000***
Cost Frying Pan	-0.249	0.200	-1.245	0.216
F-Value	24.481			
R^2	0.507			
R^{-2}	0.486			

Source: Field, survey 2019, ***** indicates coefficient significant at 1% level.**

https://dx.doi.org/10.22161/ijeab.51.18 *ISSN: 2456-1878*

The coefficients of cost purchasing (1.431), cost of labour (0.479) and cost of firewood (6.077) were significant at 1% level. This implies that an increase in unit of cost of purchasing, cost of labour and cost of fire wood will affect cashew processing by 1.431, 0.479 and 6.077 respectively. However the coefficient of cost of frying pan, and cost of processing were not significant and therefore do not significantly influence cashew processing in the study area.

IV. CONCLUSION AND RECOMMENDATION

The study was on economics of cashew processing in Gwer East Local Government Area of Benue State, Nigeria. The study found that majority of the processors was female in their productive age, married, considerable education level, and vast years of experience. From the results of costs and returns of cashew processing, the enterprise is profitable in the study area due to the positive values of the average gross margin.

The study also revealed certain factors that influenced cashew processing in the study area to be cost of purchasing, cost of fire wood and cost of labour. These have coefficients that were found to be significant in cashew processing in the study area.

The study recommends the provision of inputs facilities needed by processors by Government agencies and non-governmental organizations so as to reduce the cost incurred in processing of cashew in order to increase the profitability of cashew processing in the study area.

Processors and other individuals should come together to form cooperatives that will enable them access credit from banks and other financial institutions that will help in efficient production. Processors of cashew nut should seek for more improved production practices so as to improve their level of production to obtain even higher profits. Male folks are also encouraged to be involved in the business since it is profitable.

REFERENCES

[1] Hammed, L.A and Anikwe, J.C. (2008). Cashew Nuts and Production Development in Nigeria. *Journal of Scientific Research, 3(1):54-61*.

[2] Salau, S.A., Popoola, G.O. and Nofiu.B. N (2017). Analysis of Cashew Nuts Marketing in Kwara State, Nigeria. Federal University Oye Ekiti, Ekiti State, Nigeria (*FUOYE) Journal of Agriculture and Human Ecology*1(1): 34-44,

[3] Azam-Ali, S.H and Judge, E.C. (2001) ITDG Small Scale Cashew Nut Processing Schumacher Center for Technology and Development, Bourton on Dunsmore, Rugby, Warwickshire, UK pp 70

[4] Fumnanya, A (2018) Available on Venturesafrica.com/Nigeria-is-the-4th-largest-producer-of-cashew-in-the-world/ retrieved on 25/10/2019

[5] Opayemi, O. (2012) Proposal for New Waste Management System in Nigeria Lagos State, in Oladipupo, S., Lalita, S and Samuel, O. (2016) Empirical Investigation of Formal and Informal Sectors in Waste Recycling of the Municipal Waste Management System of Developing Countries, The Case Study of Lagos State, Nigeria *Journal of Environment and Ecology* 7(2):1-13

[6] Farayola, C.O; Akintonde, J.O.; Awoyemi, S.O and Akintaro, O.S. (2013). Economic Analysis of Cashew Nut Marketing Among Produce Buyers in Ogbomoso Metropolis of Oyo State, Nigeria. *International Journal of Agriculture Innovations and Research, 2(1):130-136.*

[7] Jaffee, S and Morton, S (1995) Private Sector High- Value Food Processing and Marketing a Synthesis of African Experience in Jaffee, S and Morton, J (eds), Marketing Africa's High Value Foods: Comparative Experience of an Emergent Private Sector, Dubuque: Kendall/ Hunt Publishing Company.

[8] Food and Agriculture Organisation (2001) Bulletin.

[9] Uwagboe E, Obatolu B. and Adeogun S. (2006). Information Delivery and its Effects on Production of Cashews in Oyo State. In: Proceedings of the Farm Management Association of Nigeria (FAMAN), held at Forestry Research Institute of Nigeria, Federal College of Forestry, Plateau State. (Eds.)

[10] Pascal, B. N.M (2013) Analysis of Socila Economic Factors Affecting Cashewnut Production in Ruangwa District, Tanzania. Published M.Sc. Dissertation, Department of Economics, University of Mzumbe, Tanzania pp 93 Retrieved on 15th October, 2019

[11] Enwelu, I.A., Ugwu, S.T., Ayogu, C.J and Ogbonna, O.I, (2014) Gender Roles and Challenges of Small Scale Cashew Nut Processing Enterpris in Enugu North, Nigeria. Journal of Educational and Social Research. 4(7): 1-6

[12] Samaripitha, A. Vasudev, N. and Suhasini, K. (2016) Socio-economic Characteristics of Rice Farmers in combined states of Andhra Pradesh Asian *Journal of Agricultural Extension, Economics and Sociology* 13(1): 1-9

[13] Sirela, B., Sarawgi, A.K., and Yogeshwari, S. (2018) Economic Analysis of Cashew Nut Processing Units in Srikakulam District of Andhra Pradesh, India *International Journal of Current Microbiology and Applied Sciences* 7(11): 195-202

https://dx.doi.org/10.22161/ijeab.51.19 *ISSN: 2456-1878*

Variability, Heritability, and Genetic progress of Maize Population F2 as a result of crossing BSM0729S3-A with BAP 27799-1

Supawi[1], Irfan Suliansyah[2,] Aprizal Zainal[3]

[1]Department of Agronomy, Faculty of Agriculture, Andalas University, Padang, Indonesia
Email: awietimpeh@gmail.com
[2]Professor in Plant Physiology, Department of Agrotechnology, Faculty of Agriculture, Andalas University, Indonesia
[3]Associate Professor, Department of Agrotechnology, Faculty of Agriculture, Andalas University, Indonesia

Abstract— *The research aims to see the variability value, heritability and genetic progress of the F2 maize population which is the result of crossing BSM0729S3-A with BAP27799-1. The final goal of this research series is to assemble high yielding composite maize. The study was conducted from April to July 2019 on the community fields, Nagari Sitiung, Sitiung District, Dharmasraya District, West Sumatra. The material used was maize seed population F2 (the result of crossing BSM0729S3-A with BAP27799-1) and the two elders were BSM0729S3-A and BAP27799-1. The observed variables were plant height, age of anthesis, age of hair appearing, cob weight without maize husk, cob length, cob diameter, and seed weight per cob. The results of the analysis of the variability values showed that all the observed variables had a wide variability, whereas based on the results of the analysis of the heritability values showed that (except for the length of the cob) all the observed variables had high heritability values. Based on the results of the analysis of the genetic progress value, the plant height variable, the cob weight without maize husk, the diameter of the cob, and the weight of the cobs of seed have a high genetic progress value, the anthesis age variable and the age of hair appearing have a high genetic progress value, while the length of the cob variable has a value low genetic progress. It can be concluded that the selection activities in the F2 population to get high-yielding maize seeds can be carried out effectively and efficiently.*

Keywords— ***variability, heritability, genetic progress, maize population F2.***

I. INTRODUCTION

Maize (*Zea mays*) is one of the economically valuable cereals and has the opportunity to be developed in Indonesia. This commodity is very multipurpose, besides using as food, it is also used for feed, as well as industrial raw materials. In fact, domestic maize production has not been able to meet their needs so they have to import. According to data from the Central Statistics Agency (BPS), the volume of maize imports as of September 2018 is 477 thousand tons [1]. One of the causes of Indonesia's national maize supply shortage is due to low productivity.

One of the efforts to increase maize productivity can be done by focusing on the components of production technology, namely superior varieties both hybrid and free-range. Both types of these varieties have advantages and disadvantages. Hybrid varieties have high yield potential but must be cultivated in fertile areas and more intensive maintenance, meanwhile free extract maize has wider adaptability, cheap seed prices, and the seeds can be used directly in the next planting season, but the level of production lower than hybrid maize. Because the productivity of composite maize is still low, efforts are needed to improve the population by doing selection in order to get high-yielding composite maize varieties.

Composite maize is maize originating from a population of random crosses (at least 5 times) from mixing seeds of the same amount from several varieties of free-lined, synthetic or hybrid. Selecting the F2 generation period for a number of cross-combination results in the context of the formation of high-yield (seed) composite maize is needed. To ensure a series of activities in the framework of producing composite maize, the calculation of the value of variability, heritability and genetic progress is very necessary. The purpose of this study was to look at the variability value, heritability and genetic progress of maize population F2 which was the result of the crossing of BSM0729S3-A with BAP27799-1.

II. MATERIALS AND METHODS

2.1 Implementation Research

The study was conducted for four months from April to July 2019. The research was carried out in the

https://dx.doi.org/10.22161/ijeab.51.19 *ISSN: 2456-1878*

community fields in Dharmasraya Regency, the location of the research location is at coordinates 1 ° 00′ 36.78" LS and 101 ° 37′ 30.57" Administratively, this area is included in the area of Sitiung Subdistrict, Dharmasraya Regency, West Sumatra Province. The materials used were population F2 maize seeds (crossing results of BSM0729S3-A with BAP27799-1) and the two elders were BSM0729S3-A and BAP27799-1, Urea fertilizer, SP-36, and KCl, Ridomil 35 SD, Furadan, Herbicide, and Insecticide. The tools used were tractors, torches, hoes, gauges, calipers, digital cameras, sickles, permanent markers, digital scales, observation guidelines, and stationery.

2.2 Data analysis

The observed variables were plant height, age of anthesis, age of hair appearing, weight of cob without maize husk, and weight of seeds per cob. Observational data for each variable were analyzed for mean values, genetic variability, heritability, and estimated values of genetic progress. The average can be calculated using the following formula:

$$\mu = \frac{\Sigma Xi}{n} = \frac{X1 + X2 + X3 + \cdots + Xn}{n}$$

Notes:

μ	:	Average
ΣXi	:	Addition of figures for all data
n	:	Amount of data

Genetic variance and Standard deviation can be calculated using the formula:

$$\sigma^2 = \frac{\Sigma(xi - \mu)^2}{n - 1}$$

$$\sigma = \sqrt{\frac{\Sigma(xi - \mu)^2}{n - 1}}$$

Notes:

σ^2	:	Varian
σ	:	Standard deviation
μ	:	Average
xi	:	1st, 2nd data etc.
n	:	Amount of data

Genetic variability is said to be broad if $\sigma^2 g \geq 2$ standard deviations (sd), and said to be narrow when σ^2 g ≤ 2 standard deviations (sd) [2]. Heritability can be calculated using the formula:

$$h^2_{(BS)} = \frac{\sigma^2 F2 - \sqrt{(\sigma^2 p1)(\sigma^2 p2)}}{\sigma^2 F2} \text{ X } 100\%$$

Notes:

h^2	:	The value of heritability is broad meaning
σ^2_{F2}	:	Variability values in population F2
σ^2_{P1}	:	The value of variability in the first population
σ^2_{P2}	:	the value of variability in the second population

Criteria for heritability: Low (h2 bs <0.2); Medium (0.2 <h2 bs ≤ 0.5); High (h2 bs> 0.5).

Genetic Progress (KGH) can be calculated using the formula:

$$KGH = i.\ h^2.\ \sigma p$$

$$\%KGH = \frac{KHG}{\mu} \text{ x } 100\%$$

Notes:

KGH	:	Genetic progress
I	:	The intensity of the selection, 10% = 1.76
h^2	:	Heritability
σp	:	Phenotype standard deviation
μ	:	Average value

Criteria for genetic progress expectations according to [3]: low (0 - 3.3%); rather low (3.31% - 6.6%); quite high (6.61% - 10%); high (> 10%).

III. RESULT AND DISCUSSIONS

3.1 Value of Variability, Heritability and Genetic Progress of Plant Height Variables

Based on the analysis of plant height variables, the population of F2 from the crossing of BSM0729S3-A with BAP27799-1 has wide genetic variability (KG), high heritability, and high genetic progress (Table 1). Wide genetic variability will make effective selection activities in order to get the desired plant criteria. The effectiveness of selection will be more efficient if the estimated value of heritability (hbs2) is high [4]. Heritability is a component in calculating the value of expected genetic progress [5], where a high heritability value is accompanied by high genetic progress, of course, it will be very effective and efficient for the selection activities done

https://dx.doi.org/10.22161/ijeab.51.19 *ISSN: 2456-1878*

Table 1. Results of analysis of genetic variability, heritability and genetic progress on plant height variables F2 from the crossing between BSM0729S3-A and BAP27799-1

Plant Height (cm)	Analysis results
μ	208.77
$\sigma^2 g$	981.59
2σg	62.66
KG	Wide
H_{bs}^2	0.54
H_{bs}^2 criteria	High
%KGH	14.36
KGH criteria	High

Note: μ = average; $\sigma^2 g$ = genetic variability; 2σg = standard deviation; KG = genetic variability; H_{bs}^2 = heritability.

Plant height is one of the agronomic characters which is usually the higher a plant means the use of sunlight for photosynthesis will be optimal, so that it will produce optimal fruit as well. In research [6] states that the production is positive correlated significantly with plant height. So the optimal plant height will produce optimal production as well.

3.2 Values of Variability, Heritability and Genetic Progress Variable anthesis and hair age

Based on the analysis of the anthesis age and the age at which hair appeared, the F2 population resulting from crossing of BSM0729S3-A with BAP27799-1 had wide genetic variability (KG), high heritability, and high genetic progress (Table 2 and Table 3).

Table 2. The results of the analysis of the value of genetic variability, heritability and genetic progress on the age anthesis population F2 from the crossing between BSM0729S3-A and BAP27799-1

Age of anthesis	Analysis results
μ	55.82
$\sigma^2 g$	5.59
2σg	4.73
KG	Wide
H_{bs}^2	0.91
H_{bs}^2 criteria	High
%KGH	6.81
KGH criteria	High enough

Note: μ = average; $\sigma^2 g$ = genetic variability; 2σg = standard deviation; KG = genetic variability; H_{bs}^2 = heritability.

Table 3. The results of the analysis of genetic variability, heritability and genetic progress on the age variables appearing hair F2 population results from crossing BSM0729S3-A with BAP27799-1

Age of hair appears	Analysis results
μ	57.43
$\sigma^2 g$	7.72
2σg	5.56
KG	Wide
H_{bs}^2	0.95
H_{bs}^2 criteria	High
%KGH	8.10
KGH criteria	High enough

Note: μ = average; $\sigma^2 g$ = genetic variability; 2σg = standard deviation; KG = genetic variability; H_{bs}^2 = heritability.

Usually flowering age is positively correlated with age of harvest. If the age of flowering is fast then the age of harvest will also be fast. As stated by [7] that in rice plants whose flowering age is faster has a faster generative phase too, so that the faster the flowering plants the faster the harvesting time.

3.3 Values of Variability, Heritability and Genetic Progress Variable weight of cob without maize hudk and seed weight per cob.

Based on the analysis of the weightless cob variable and seed weight per cob, the population of F2 from the crossing of BSM0729S3-A with BAP27799-1 has wide genetic variability (KG), high heritability, and high genetic progress (Table 4 and Table 5).

Table 4. Results of analysis of genetic variability, heritability and genetic progress in the weight variable of cob without maize husk population F2 resulting from crossing of BSM0729S3-A with BAP27799-1

cob weights without maizehusk	Analysis results
μ	128.38
$\sigma^2 g$	2343.42
2σg	96.82
KG	Wide
H_{bs}^2	0.81
H_{bs}^2 criteria	High
%KGH	53.47
KGH criteria	High

Note: μ = average; $\sigma^2 g$ = genetic variability; 2σg = standard deviation; KG = genetic variability; H_{bs}^2 = heritability.

https://dx.doi.org/10.22161/ijeab.51.19 *ISSN: 2456-1878*

Table 5. The results of the analysis of genetic variability, heritability and genetic progress on seed weight per cob F2 population resulted from crossing BSM0729S3-A with BAP27799-1

Seed weight per cob	Analysis results
μ	85.12
σ^2g	1140.48
2σg	67.54
KG	Wide
H_{bs}^2	0.75
H_{bs}^2 criteria	High
%KGH	52.29
KGH criteria	High

Note: μ = average; σ^2g = genetic variability; 2σg = standard deviation; KG = genetic variability; H_{bs}^2 = heritability.

The wide genetic variability caused by the population used is F2 seed with the highest level of segregation. Wide genetic variability will make the effective selection process in order to get the desired plant criteria, namely maize plants with high yield of seeds. Characters that have wide genetic variability and are accompanied by high heritability values will accelerate the selection process for the characters developed. According to [8] heritability is needed to determine the extent to which the appearance of a plant character is influenced by genetic and environmental factors. If the heritability is high, most of the phenotypic variation is caused by genetic variation, so the selection will get genetic progress [9].

IV. CONCLUSION

Selection activities in the F2 generation resulting from crossing BSM0729S3-A with BAP27799-1 can be effective because the observed variables, generally have wide genetic variability, quite high tilll high heritability and high genetic progress.

REFERENCES

[1] BPS. 2018. Buletin Statistik Perdagangan Luar Negeri : Impor. Badan Pusat Statistik, Jakarta.

[2] Wardani, S., D. Wirnas, dan Y. Wahyu. 2015. Seleksi Segregan Gandum (*Triticum aestivum* L.) pada Dataran Tinggi. *Jurnal Agronomi Indonesia*. 43 (1) : 45-51.

[3] Suprapto, dan N. M. Kairudin. 2007. Variasi Genetik, Heritabilitas, Tindak Gen dan Kemajuan Genetik Kedelai (*Glycine max Merrill*) pada Ultisol. *Jurnal Ilmu-Ilmu Pertanian Indonesia*. 9 (2): 183-190.

[4] Moedjiono, M. J. Mejaya. 1994. Variabilitas Genetik Beberapa Karakter Plasma Nutfah Jagung Koleksi Balittas Malang. *Zuriat.* 5(2):27-32.

[5] Koppad, S. B., Chavan, M. L., Hallur, R. H., Rathod, V. and Shantappa, T. 2015. Variability and Character Association Studies in Ridge Gourd (*Luffa acutangula Roxb*.) With Reference to Yield Attributes. *Journal of Global Biosciences*. 4(5) : 2332-2342.

[6] Salamah-Umi, W. B. Suwarno, H. Aswidinnoor & A. Nindita. 2017. Keragaan agronomi dan potensi hasil genotype jagung (*Zea mays* L.) generasi S1 dan S2 di dua lokasi. J. Agronomi Indonesia. 45(2): 138-145.

[7] Chandrasari, S.E., Nasrullah, Sutardi. 2013. Uji daya hasil delapan galur harapan padi di sawah. Vegetalika 1:99-107.

[8] Ayalneh, T., Z. Habtamu, A. Amsaluy. 2012. Genetic Variability, Heritability and advance in tef (*Eragrotis tef* (Zucc.) Trotter) lines at Sinana and Adaba. *International Journal of Plant Breeding*. 6 (1): 40-46.

[9] Suprapto, dan N. M. Kairudin. 2007. Variasi Genetik, Heritabilitas, Tindak Gen dan Kemajuan Genetik Kedelai (*Glycine max Merrill*) pada Ultisol. *Jurnal Ilmu-Ilmu Pertanian Indonesia.* 9 (2): 183-190.

https://dx.doi.org/10.22161/ijeab.51.20 ISSN: 2456-1878

In-vitro screening of Iranian Landraces (Triticum*aestivum* L.) at the seedling stage for water stress tolerance using Polyethylene glycol (PEG 6000)

Amandeep Kaur[1]*, Rashpal Singh Sarlach[2]

[1]Department of Botany, Punjab Agricultural University, Ludhiana, India
[2]Department of Plant Breeding & Genetics, Punjab Agricultural University, Ludhiana, India
*Corresponding author:deepaman3305@gmail.com

Abstract— *To understand, the parameters in wheat which can be used as criteria for drought tolerance, the present investigation was carried out on eight genotypes to an optimum dose of Polyethylene glycol (PEG6000) for water stress tolerance at germination and seedling stage. The experiment was conducted in the Department of Plant Breeding and Genetics with three replications under completely Randomized Design,7 different concentration of Polyethylene glycol that were 0, 6%, 8%, 10%,12%, 14% 16% and 18% used in this experiment which were prepared according to weight by volume method. There was a significant reduction in seedling parameters with an increasing concentration of Polyethylene glycol. Based upon the vigor index low treatment of polyethylene glycol decreased the seedling parameters to a lesser extent than high concentration. The maximum difference in all seedling parameters was observed at 14% but as the concentration of polyethylene glycol increased (16% and 18%) there was a greater reduction in the root, shoot length and vigor index as compared to 14% of Polyethylene glycol which had an intermediate effect on seedling parameters. Identifying the drought-tolerant wheat genotypes during the seedling stage by in-vitro screening is a physiological approach which assists breeder for rapid selection of genotypes.*

Highlights

- *Reduction in seedling parameters with increasing concentration of Polyethylene glycol*
- *Water stress-tolerant landraces have maximum vigor index*
- *For in-vitro screening, the moderate concentration of Polyethylene glycol should be used*

Keywords— ***Water stress, Polyethylene glycol, Iranian wheat landraces, screening, and seedling growth.***

I. INTRODUCTION

Wheat is the staple food contributing 20% calories to the world's population, with a total harvest area of 2.1 million km^2 and global production of 700 million tonnes (Shiferawet al2013 and Lobel and Gourdji 2012). Water deficit is one of the major constraints in agricultural production including wheat, which has devastated the economy of many countries. Water is also becoming a scarce commodity and its severity has been forecast further in this area as well, in the years to come. Water stress affect plant at any stage but germination and seedling growth are severely affected however these traits plays a vital role in determining yield (Raufet al 2007). Roots are more sensitive to drought stress because regulation of water mechanism occurs inside the apoplast of the root (Ghafoor 2013).Root and shoot length are important traits for screening water tolerance in the selection of drought tolerance at the early seedling stage is frequently accomplished by using drought-induced chemical Polyethylene glycol (PEG 6000).Underwater stress osmotic adjustment is very necessary for plants to cope up with stress. Polyethylene glycol (PEG 6000) is a neutral osmotically group of polymers having high molecular weight used to induce water deficit in plants by modifying the osmotic potential of nutrient solution in a controlled manner. Screening of seedling traits using PEG is the most simple and cost-effective method for screening maximum germplasm. Identification of wheat genotypes that can tolerate water stress conditions is vital to boost wheat production and also helpful for understanding yield-limiting factors. To date, most of the Iranian germplasm has not been characterized and used in plant breeding

https://dx.doi.org/10.22161/ijeab.51.20 *ISSN: 2456-1878*

(Hoisington et al 2010). Therefore assessing genetic variation and differentiation of Iranian wheat landraces and cultivars will facilitate the effective use of these valuable genetic resources in future breeding to broaden genetic diversity. The present study was carried to pinpoints the optimum dose of Polyethylene glycol 6000 to create water stress at the seedling stage so that the response of Iranian landraces for stress tolerance elucidate and landraces screened efficiently.

II. MATERIALS AND METHODS

The experiment was carried out in a completely randomized design with three replications and plant material was procured from International Maize and Wheat Improvement (CIMMYT). The study was carried out in the Research Laboratory of wheat section Plant Breeding and Genetics, Punjab Agricultural University Ludhiana. Five randomly selected Iranian landraces along with 3 checks were included in the standardization experiment. The solution of Polyethylene glycol (PEG 6000) was prepared according to weight by volume method (Bayoumi et al 2008). The different concentration of Polyethylene glycol (PEG 6000) (6%.8%,10%,12%,14%,16% and 18%) was prepared in the present study. To prepare T1 (6%) 60 g of Polyethylene glycol (PEG 6000) was dissolved in distilled water to raise the volume to one liter and similarly all treatment T2 (8%) 80g, T3 (10%) 100g, T4 (12%) 120g, T5 (14%) 140g, T6 (16%) 160g, T7 (18%) 180g of Polyethylene glycol (PEG 6000) was dissolved in distilled water to raise the volume to one liter. Polyethylene glycol(PEG 6000) was used to induce drought stress during the germination and seedling stage by using a cigar roll method (Zhu et al2005). After surface sterilization of seeds with 0.1 % mercuric chloride solution for 2 minutes, the seeds were washed with water and kept for germination on germination paper in distilled water (control) and PEG 6000 solution of different concentrations(6,%8%,10%,12%,14%,16%, and 18%) for water-stress studies. Rolls were placed in a growth chamber in darkness at 25°C for 3 days under a photoperiod of 16/8 hours. This experiment was conducted on check varieties to standardize the concentration of Polyethylene glycol (PEG 6000) which affects germination and seedling growth The data on germination percent, root length, shoot length, root fresh weight, shoot fresh weight, root dry weight, shoot dry weight was recorded from different treatments and vigor index was calculated by using formula

Vigor index= Germination percentage× seedling dry weight (Root+ shoot dry weight in g)

III. RESULTS AND DISCUSSIONS

Analysis of Variance

Analysis of variance for all the seedling traits for water stress tolerance was conducted. ANOVA revealed significant differences among cultivars water stress and the interaction effect between the landraces and Polyethylene glycol (PEG) concentration. The mean square due to treatment, genotype, and interaction between treatment and genotype were highly significant for all the seedling traits. (Table 2)

Germination percentage

Maximum germination percentage (100%) was observed under control conditions in Iranian landraces and commercial check varieties followed by (96.2) in T1 and (90.4) in T2 were 6% and 8% PEG applied respectively(Table 1). With the increasing concentration of Polyethylene glycol (PEG6000), there was a decrease in the rate of germination. Minimum germination percentage i.e. was 35.5 and 24.7 recorded at 16% and 18% of peg 6000. Iranian landraces and wheat cultivars showed 70% germination at 140 g/l Polyethylene glycol (Table 1).A similar result of the reduction in germination percentage with increasing peg concentration was reported by Ghodsi (2004) and Rauf et al (2007).

Root length

Mean values regarding root length are presented in (Table 1). The root length decreased significantly with increasing moisture stress in Iranian landraces and wheat cultivars. Maximum root length (21.0) was observed under control conditions followed by 17.9 and 17 in T1 and T2 respectively whereas minimum (3.35) root length was recorded at 18%. Root length at T4 and T5 showed a difference of 1.4 cm (Table 1). The result of the present study consisted of the findings of Rauf et al (2007), Singh et al (1994) and Jajarmi et al (2009).

Shoot length

With increasing stress levels there was a reduction in shoot length in Iranian landraces and wheat cultivar. The decrease in shoot length was maximum at 18% Polyethylene glycol (4.86 cm) followed by 16% (6.88 cm) and 14% (11.0) peg respectively (Table 1). Under control condition, maximum shoot length was recorded that was 17.4 cm followed by T1 (15.3 cm)and T2(14.2 cm) respectively. (Table 1).Reduction in shoot length was similar to the findings of Kamran et al (2009).

Root and shoot fresh weight

Maximum root (0.13g) and shoot (0.30g) fresh weight were recorded in control conditions followed by T1and T2 respectively whereas minimum root fresh weight was recorded at 16% and 18% that was 0.05 respectively

https://dx.doi.org/10.22161/ijeab.51.20 *ISSN: 2456-1878*

(Table 1). A reduction of 0.04g was observed in T5 (14%) as compared to T4 (12%) peg (Table 1). Minimum shoot fresh weight was observed at 18% peg (0.03) followed by 14% peg (0.06) respectively. There was a reduction of 0.03g and 0.04 in T4 (12%) and T5 (14%) as compared to T3 (10%) (Table 1). The result of the present is similar to the findings of Bayoumi et al (2008) who reported that there was a reduction in root and shoot biomass with increasing concentration of Polyethylene glycol

Root and shoot dry weight:

Maximum root dry weight (0.05g) was recorded in control conditions, T2, T3, and T4, whereas in T7 (18%) minimum root dry weight (0.02g) was observed. 0.04 g was recorded at 14% peg followed by (0.03) in 12% of peg (Table 1). The maximum and minimum shoot dry weight wasrecorded in all treatments were 0.03 and 0.02 g respectively. The decreasing trend in shoot and root dry weight with increasing concentration of polyethylene glycol were reported by Kamran et al (2009) and Ahmad et al (2013) in wheat which is consistent with the present study.

Vigor index

The highest value (3724) of seedling vigor was observed in control conditions in Iranian landraces and commercial check varieties followed by 3063 at 6% peg and 2734 at 8% peg respectively, however, the seedling vigor decrease with increasing concentration of polyethylene glycol (Table 1). The least value of vigor index was recorded at 18% peg that was 290. There was a difference of 140 and 746 in T3 and T4 as compared to the vigor index in T2 (2754). There was a reduction in vigor index with increasing concentration of Polyethylene glycol (PEG) as reported by Cokkizgin (2013) in pea, the similar result had been reported by Moras et al (2005) in the bean.Reduction in vigor index inwheat with increasing concentration of Polyethylene glycol (PEG 6000) reported by Tamiru and Ashagrerin (2014).

Correlation studies

The correlation coefficient was calculated among all the characters (Table 3). Germination percentage showed a positive relation with root length, shoot length,root fresh weight, shoot fresh weight and seedling vigor. It means an increase in germination percentage would also increase in these traits. Root length is strongly correlated with shoot length and vigor index. These results are following the findings of Khan et al (2002). Correlation analysis showed that shoot length exhibited a positive association with root length and vigor index which means an increase in shoot length will also cause an increase in these parameters. These results are supported by Akram et al (1998). Root and Shoot fresh weight were correlated with root length whereas the vigor index showed a strong positive correlation with root length and shoot length.

IV. CONCLUSION

Generally from the present study, it is concluded that there is a reduction in germination and seedling growth with increasing concentration of polyethylene glycol. The low treatment of Polyethylene glycol (, 6% and 8%) decreased the seedling parameter to a lesser extent than moderate treatment (10%,12%, and 14%) and high concentration (16% and 18%) of Polyethylene glycol. Germination percentage, root length, shoot length and vigor index of wheat genotypes showed lesser reduction till 12% of Polyethylene glycol. The maximum difference in all seedling parameters observed at 14% of Polyethylene glycol while at higher concentration of polyethylene glycol there was a major reduction in germination percentage, root length, shoot length and vigor index of wheat cultivars. Based upon the vigor index all the genotypes respond better at 14% of Polyethylene glycol (PEG 6000). Vigor index can increase the crops seasonal water use efficiency by as much as 25% and is recognized as a trait to select for improving yield under water stress (Richards et al 2002) and Botwright et al (2002). The genotypes which have higher seedling vigor index also have better germination percentage, root length and shoot length (Hafid et al 1998)

Table 1: Effect of different drought levels on growth parameters on wheat genotypes

Parameters	Peg(g/l)	IWA 860005	IWA 8600303	IWA 8600542	IWA 8606753	IWA 8600824	C-306	PBW175	PBW660	Mean	LSD
Germination Percentage	0%	100	100	100	100	100	100	100	100	100a	6.23
	6%	90	100	100	100	90	100	90	100	96.25b	
	8%	86.5	90.5	90.5	90.5	90	100	85.5	90	90.44c	
	10%	80.5	85.5	90.5	85.5	85.5	96.6	85.5	95.5	88.14d	
	12%	80	83.3	85.5	80	80	95.5	80	85.5	83.73e	
	14%	76.5	80.5	80	70.5	70	85.5	80	85.5	78.56f	

	16%	40.5	7.05	60.5	40.5	4.05	40	50.5	40.5	35.5g	
	18%	20.5	5.5	40.5	20	20.5	40	20	30.5	24.7h	
Root Length	0%	18.5	19	18	20.5	20.5	23.5	23.5	24.5	21a	1.33
	6%	17.5	15.5	18	15.5	18	20.5	17.5	21	17.9a	
	8%	17	14	15.5	14	17.5	19	19.5	19.5	17 a	
	10%	16.5	14	14	13.8	15.5	19	17.5	17.5	16.0 a	
	12%	15	13.5	12.5	12	14	16	12	15.5	13. b	
	14%	10.8	12	12	7.8	13.5	16.3	12	15	12. b	
	16%	6.5	9.8	7.8	4.5	4.5	7.8	9.8	7.8	7.31 c	
	18%	3.5	2.5	2.5	3.5	3.5	4.5	4.5	2.3	3.35d	
Shoot Length	0%	15	17	15	17	18.5	19.4	19	18.5	17.4a	1.79
	6%	15.5	13.9	18	13.5	14.5	15.5	15.5	16	15.3b	
	8%	15	13	12.5	12.8	14	16	14.5	15.5	14.2b	
	10%	13.5	13.7	12	13	13	14.5	14	15	13.6c	
	12%	14	12	12	11	12	15	12	14	12.8d	
	14%	7.8	11	9.8	7.5	11.5	15	12	13.5	11.0e	
	16%	5	7.5	6.5	4	4.5	9.5	7.5	10.5	6.88f	
	18%	5.5	3	3	2.9	4	7	5	8.5	4.86g	
Root Fresh weight	0%	0.12	0.14	0.13	0.12	0.12	0.13	0.14	0.14	0.14a	0.44
	6%	0.14	0.12	0.1	0.13	0.16	0.15	0.15	0.16	0.13a	
	8%	0.13	0.12	0.12	0.13	0.14	0.12	0.13	0.13	0.13a	
	10%	0.1	0.1	0.11	0.09	0.12	0.12	0.09	0.12	0.11a	
	12%	0.09	0.08	0.08	0.1	0.1	0.19	0.08	0.08	0.1a	
	14%	0.06	0.06	0.07	0.08	0.06	0.06	0.05	0.04	0.06a	
	16%	0.04	0.05	0.06	0.05	0.04	0.04	0.05	0.04	0.05a	
	18%	0.04	0.05	0.06	0.05	0.04	0.04	0.05	0.04	0.05a	
Shoot Fresh weight	0%	0.37	0.3	0.29	0.2	0.18	0.41	0.38	0.28	0.30a	0.48
	6%	0.24	0.25	0.22	0.18	0.18	0.16	0.2	0.2	0.20a	
	8%	0.15	0.14	0.16	0.16	0.14	0.15	0.15	0.14	0.15a	
	10%	0.1	0.12	0.14	0.1	0.1	0.11	0.09	0.08	0.11a	
	12%	0.1	0.07	0.06	0.08	0.09	0.08	0.08	0.07	0.08a	
	14%	0.07	0.06	0.09	0.08	0.06	0.06	0.05	0.05	0.07a	
	16%	0.06	0.05	0.08	0.06	0.06	0.05	0.04	0.04	0.06a	
	18%	0.05	0.03	0.02	0.02	0.03	0.02	0.02	0.03	0.03a	
Root Dry weight	0%	0.05	0.07	0.04	0.06	0.05	0.06	0.03	0.04	0.05a	0.30
	6%	0.05	0.02	0.04	0.06	0.05	0.03	0.04	0.06	0.04a	
	8%	0.05	0.05	0.06	0.04	0.05	0.03	0.05	0.05	0.05a	
	10%	0.05	0.06	0.03	0.03	0.05	0.04	0.05	0.05	0.05a	
	12%	0.04	0.05	0.04	0.05	0.07	0.05	0.04	0.05	0.05a	
	14%	0.06	0.07	0.04	0.05	0.03	0.03	0.04	0.03	0.04a	
	16%	0.03	0.03	0.03	0.03	0.02	0.03	0.03	0.04	0.03a	
	18%	0.04	0.02	0.03	0.02	0.01	0.02	0.02	0.01	0.02a	
Shoot dry weight	0%	0.04	0.03	0.04	0.03	0.04	0.03	0.03	0.03	0.03a	0.19
	6%	0.02	0.03	0.02	0.03	0.02	0.03	0.02	0.02	0.02a	

	8%	0.02	0.03	0.02	0.02	0.02	0.02	0.03	0.03	0.02a	
	10%	0.02	0.02	0.02	0.03	0.03	0.02	0.02	0.04	0.03a	
	12%	0.04	0.03	0.02	0.03	0.02	0.02	0.02	0.02	0.03a	
	14%	0.02	0.03	0.03	0.02	0.03	0.03	0.03	0.03	0.03a	
	16%	0.02	0.03	0.03	0.04	0.03	0.02	0.03	0.02	0.03a	
	18%	0.03	0.02	0.01	0.02	0.01	0.01	0.01	0.01	0.02a	
Vigor Index	0%	3350	3600	3400	3200	3400	4290	4250	4300	3724a	338.4
	6%	2970	2940	3300	2900	2925	3600	2970	2900	3063b	
	8%	2768	2443	2400	2400	2300	3500	2907	3150	2734c	
	10%	2415	2368	2353	2291	2291	3236	2693	3103	2594d	
	12%	2320	2124	2094	2040	2080	1920	1900	1422	1988e	
	14%	1851	1744	1078	1750	2565	1920	1900	465	1659f	
	16%	1219	865	344	364.5	550	692	684.5	184.5	613g	
	18%	277	222	128	153	460	440	329	310.5	290h	

Values in mean column sharing the same letter are statistically non- significant at 5%

Table.2: Analysis of variance for seedling traits of Iranian landraces and check varieties under different drought level

Source of variation	DF	Germ%	RL	SL	RFW	SFW	RDW	SDW	VI
Genotypes	7	531.4*	51.9*	35.04*	0.123*	0.476*	0.345*	0.639*	1037059*
Drought levels	7	17727.3*	817.53*	386.7*	0.375*	0.141*	0.252*	0.777*	323295*
Cultivars× Drought levels	49	153.96*	7.011*	11.07*	0.797*	0.93*	0.378*	0.145*	45374*
Error	128	44.04*	1.01*	0.863*	0.255*	0.229*	0.13*	0.364*	13020.4*
Total	191								
Coefficient of variation		8.85	7.35	7.69	5.56	4.01	8.47	7.42	17.74

Abbreviations- germ%- Germination percentage, RL- Root length, SL- Shoot length, RFW- Root fresh weight, SFW- Shoot fresh weight, RDW- Root dry weight, SDW- Shoot dry weight and VI- Vigor index *Significant at 5% level

Table 3: Correlation coefficient among various seedling traits of different wheat genotypes

	GP	RL	SL	SFW	RFW	RDW	SDW	VI
GP	1	0.87**	0.86**	0.89**	0.88**	0.53**	0.35**	0.85**
RL		1	0.93**	0.68**	0.82**	0.52**	0.43**	0.91**
SL		0.89**	1	0.67**	0.37**	0.43**	0.42**	0.88**
SFW		0.89**	0.67**	1	0.68**	0.43**	0.32**	0.76**
RFW		0.88**	0.37**	0.68**	1	0.5	0.18**	0.82**
RDW		0.64**	0.43**	0.4	0.5**	1	0.15**	0.48**
SDW		0.6**	0.42**	0.32**	0.18**	0.15**	1	0.34**
VI		0.89**	0.87**	0.76**	0.82**	0.48**	0.34**	1

Abbreviations- GP Germination percentage RL- Root length, SL- Shoot length, RFW- Root fresh weight, SFW- Shoot fresh weight, RDW- Root dry weight, SDW- Shoot dry weight and VI- Vigor index significant at 0.01**

REFERENCES

[1] Akram, H. M., Ahmed,N., Al,i A .,and Yar, A. 1998.Effect of stress on germination and seedling growth of wheat cultivars. *J Agri Res* **36** (3): 217-22.

[2] Bayoumi,T .Y., Ei,d M. H., and Metwali, E.M. 2008. Application of physiological and biochemical indices as.screening technique for drought tolerance in wheat genotypes. *AfrJ Biotechnol***7**: 2341-52

[3] Bortwright, T. L., Condon, A. .G., Rebetzke, G. J., and Richards, R.A. 2002. Field evaluation of early vigor for genetic improvement of grain yield in wheat. *Aust J AgricRes***53**: 1137-45

[4] Cokkizgin, A. 2013.Effects of hydro and osmo-priming on seed vigor of pea (Pisumsativum L.)*Agric Forestry Fish***2**(6): 225-28.

[5] Ghafoor, A. 2013. Unveiling the mess of red pottage through gel electrophoresis: a robust and reliable method to identify Vicia sativa and Lens culinaris from a mixed lot of split red dal. *Pak J Bot***45**: 1551-56.

[6] Ghods, M. 2004. 'Eco-physiological aspects of water deficit on growth and development of wheat cultivars'.Ph.D. Thesis, University of Tehran, Iran.

[7] Hafid, R. E., Smith, D. H., Karrou, M., and Samir, K. 1998.Physiological responses of spring durum wheat cultivars to early season drought in a Mediterranean environment.*Annals Bot***81**: 363-70.

[8] Hoisington, D., Khairallah, M., Reeves, T., Ribaut, J. M., Skovmand, B., and Taba, S. 1999.Plant genetic resources: what can they contribute toward increased crop productivity? *ProcNatlAcadSci* USA **96**: 5937-43.

[9] Jajarmi, V. 2009.Effect of water stress on germination indices in seven wheat cultivar.*World AcadSciEngTechnol***49**:105-06.

[10] Kamran, M. S., Muhammad, A., Muhammad, A .,and Nudrat, A. 2009. Alleviation of drought-induced adverse effects in spring wheat (*Triticumaestivum* L.) using proline as a pre-sowing seed treatment. *PakJ Bot***41**: 621-32.

[11] Rauf, M., Munir, M., Ul-Hassan, M., Ahmed, M., and Afzal ,M. (2007) Performance of wheat genotypes under osmotic stress at germination and early seedling growth stage. *Afr J Biotechnol***8**:971-75.

[12] Richards, R.A., Rebetzke, G. J., Condon, A.G., and VAN Herwaarden, A.F. 2002. Breeding opportunities for increasing the efficiency of water use and crop yield in temperate cereals .*Crop Sci***42**: 431-47

[13] Shiferaw, B .et al. 2013. Crops that feed the world . Past successes and future challenges to the role played by wheat in global food security. Food Security 5:291–317 .

[14] Singh, G., Bhalla, V., and Singh, G. 1994 .Differential response of wheat genotypes to moisture stress for seed germination and early seedling growth.*Indian J Agric Res***28**: 99-104.

[15] Tamiru, and Ashagre. 2014. In vitro evaluation of wheat cultivars for moisture stress. *Int J Agril Res InnovTech***4**(2): 55-60.

[16] Zhu .J.,Kaeppler. S. M., Lynch, J. P. 2005. Mapping of QTLs for lateral root branching and length in maize (Zea mays L.) under differential phosphorus supply.*Theo Appl Genet111:* 688-95.

https://dx.doi.org/10.22161/ijeab.51.21 *ISSN: 2456-1878*

The Effect of Weed Control on the Growth and Yield of Shallot (Allium ascalonicum L.)

Husni Thamrin Sebayang*, Pebrio Adi Prasetyo

Department of Agronomy, Brawijaya University, INDONESIA
*coreesponding: husni_thsby@yahoo.co.id

Abstract— *Weeds are one of several factors that cause decreased shallot production. Weed control is needed to increase production. The experiments to study the effect of weed control on the growth and yield of shallot had been conducted from June 2019 to September 2019 at Kepuharjo Village in Karangploso Sub- District, Malang Regency. The experiment used a randomized block design (RBD) with 6 treatments and 4 replications. The results showed that for treatment of weed-free, weeding at 15, 30 and 45 DAP (Days after planting), application of oxyfluorfen herbicides at a dose of 1.5 l /ha + weeding at 30 DAP, silver black plastic mulch + weeding at 30 DAP and straw mulch rice + weeding at 30 DAP the dry weight of weed significantly decreased. The growth and yield of shallot showed significantly higher with weed free treatment followed by weeding 15, 30 and 45 DAP, application of oxyfluorfen herbicide at a dose of 1.5 l/ ha + weeding at 30 DAP, silver black plastic mulch + weeding at 30 DAP and rice straw mulch + weeding at 30 DAP treatments. The growth and yield of shallot showed significantly lower with the treatment of without weed control compared with the other weed control treatments.*

Keywords— ***Shallot, Weed and Weed Control.***

I. INTRODUCTION

The shallot plant is one of the essential plants in society that functions as a spice for food and traditional medicine, and has been cultivated by farmers for a long time. The need for shallots continues to increase in line with the growing population of Indonesia, which leads to shallots having a quite high economic value. In Indonesia, the consumption of shallots tends to increase with an average growth of 8.31% kg/ capita/ year and shallot production has increased by 3.93%/ year. The increase in production was caused by an increase in harvesting area by 7.16%/ year and productivity by 1.05%/ year (Center for Data and Agriculture and Information System, Ministry of Agriculture, 2016).

Various appropriate cultivation technologies continue to be applied to increase the production of shallot plants. One of the factors that interfere the production of onion family and increase cultivation cost is the presence of weeds around plants (Vijayvergiya, 2018). Onion plants are considered as weak competitors against weeds because of their slow growth, short plant shape, shallow roots system, upright leaves and cylindrical shape making them less able to suppress the growth of weeds through the closure of plant shade (Sekara, et al., 2017). The presence of weeds can reduce crop yields because of competition for growth factors such as water, light, air, nutrients and weed also become host for pests or diseases (Bhullar, et al., 2015). Weed competition with weed can reduce onion bulbs yield by 30 - 60% (Tripathi, et al., 2013). Control of weeds on shallots needs to be performed to increase crop yields.

II. MATERIAL AND METHOD

An experiments to study the effect of weed control on the growth and yield of shallot had been conducted from June 2019 to September 2019 at Kepuharjo Village in Karangploso Sub-District, Malang Regency, at an altitude of ± 525 m above sea level and with an averages rainfall of approximately 1000 mm, average daily temperature of 14 ^{0}C and clay-type soil. The experiment used a randomized block design (RBD) consisting of 6 treatments that were repeated 4 times. The treatment of weed control are P0: without weed control, P1: weed free, P2: weeding at 15, 30 and 45 DAP (Days after planting), P3: application of oxyfluorfen herbicide with a dose of 1.5 l /ha + weeding at 30 DAP, P4: silver black plastic mulch + weeding at 30DAP and P5: rice straw mulch + weeding 30 DAP. Tillage was performed by dredging the soil with a hoe 2-3 times until the soil becomes loose. Seedbed for experimental plot were then made with placement of 2.5 m x 1.5 m, seedbed heights of 30-40 cm, the seedbed placed 50 cm apart, and the replications placed 50 cm apart. Seedlings of Tajuk shallot variety were planted with spacing of 15 x 15 cm. The basic fertilizer consists of

https://dx.doi.org/10.22161/ijeab.51.21 *ISSN: 2456-1878*

250 kg/ ha of SP 36 and 200 kg/ ha of NPK given after planting. At the age of 15 DAP, 200 kg/ ha of NPK fertilizers was given and at the age of 30 DAP, 200 kg/ ha of NPK fertilizers and 150 kg/ha of ZA fertilizer was given. Fertilizing was performed around the rows of shallot plants.Watering was performed every 2-3 days in accordance with plant conditions. Weed control in weed-free treatment was performed every 3-5 days if there are weeds that grew. Weed control with oxyfluorfen herbicide was performed using a hand sprayer at a dose 1.5 l/ ha with a water volume of 500 l/ha. Silver black plastic mulch and rice straw mulch was applied before planting. Silver black plastic mulch was perforated to grow plants. Rice straw mulch was spread with a thickness of approximately 2-3 cm, and the shallots were then planted between rice straw mulch. Weeding according to treatment was performed manually using a sickle or hoe. Observation of weed dry weight, weed control efficiency (WCE) and weed index (WI) as well as the growth and yield of shallots were carried out at 15, 30, 45 and 60 days after planting. The obtained data were analyzed using analysis of variance (F test) with a level of 5% to determine the effect of the treatment. If significant occur, the LSD (Least Significant Difference) test was carried out with a level of 5%.

Weed Control Efficiency (%)

Weed control was calculated by using the following formula (Prachand,et al., 2014):

$$WCE\ (\%) = \frac{DWC - DWT}{DWC} \times 100 \quad (1)$$

Where, WCE = Weed control efficiency (%), DWC = Dry weight of weed in control plot, DWT = Dry weight of weed in treatment plot.

Weed Index (%)

Weed index was calculated by using the following formula (Prachand,et al., 2014):

$$\text{Weed index (WI) } \% = \frac{X - Y}{X} \times 100 \quad (2)$$

Where, X = Weight of bulbs yields in treatment which highest yield, Y = Weight of bulbs yields from the treatment plot.

III. RESULT AND DISCUSSION

3.1 Weed Growth

Weed control significantly affected the weed dry weight observed at 15, 30, and 60 DAT (Table 1). The weed dry weight was significantly higher in P0 treatment (without weed control) at observations of 15, 30, 45 and 60 DAP being 0.70, 26.98, 38.63 and 63.80 g/ 0.3 x 0.4 m respectively, and significantly lower in the P1 (weed-free) treatment being 0.25, 0.38, 0.43 and 1.23 g/ 0.3 x 0.4 m respectively compared to other weed control treatments.

Table 1: Average Total Dry Weight of Weed with Various Weed Control Treatments.

Treatments	Observed weed dry weight (g/0.3 x 0.4 m) at various DAP				Observed WCE (%) at various DAP			
	15	30	45	60	15	30	45	60
P0	1.09 c (0.70)	5.21 d (26.98)	6.21 d (38.63)	7.96 d (63.80)				
P1	0.87 a (0.25)	0.93 a (0.38)	0.95 a (0.43)	1.31 a (1.23)	61.25	98.55	98.92	97.88
P2	0.95 ab (0.40)	2.81 b (7.43)	2.04 bc (3.98)	3.46 bc (11.65)	35.63	71.06	88.61	80.31
P3	0.87 a (0.25)	2.39 b (5.30)	1.83 bc (3.23)	2.60 b (6.38)	60.63	80.09	92.22	89.32
P4	0.99 ab (0.50)	3.21 bc (10.08)	1.48 ab (1.88)	3.40 bc (11.90)	27.50	60.69	94.49	80.10
P5	1.01 b (0.53)	4.05 c (17.45)	2.48 c (5.75)	4.11c (16.68)	23.13	41.09	84.84	73.72
LSD 5%	0.12	1.01	0.79	1.03				
CV	8.04	21.61	20.95	17.95				

Note: Numbers followed by the same letters for the same columns show no significant difference based on the LSD (Least Significant Difference) 5% test. CV= Coefficient of variance. DAP = days after planting. Numbers in parentheses are original numbers. Transformation √ (x + 0.5).

The dry weight of weed in the P2 (weeding at 15 DAP, 30 DAP and 45 DAP), P3 (herbicide application + weeding at 30 DAP), P4 (silver black plastic mulch + weeding at 30 DAP) and P5 (rice straw mulch + weeding at 30 DAP) treatments were significantly lower compare to without weed control. The WCE of weeds were significantly higher at P1 (weed-free) treatment being 61.25, 98.55, 98.92 and 97.88 % as observed at 15-60 dap. A research by Kumar with onion (2014) showed that the population and dry weight of weeds are significantly higher if weeds are not controlled and are lower when weed are controlled. Priya, et al. (2017) stated that oxyfluorfen herbicide is widely used by farmers at low doses and is easy to use, both pre and post-emergence and to control annual and perennial broadleaf weeds in a various field crops.

3.2 Component of Growth

Plant length did not differ between weed control treatments at 15 and 60 DAP observations and was significantly different at 30 and 45 DAP observations (Table 2).

Table 2: Average Plant Length of Shallot with Various Weed Control Treatments.

Treatments	Observed plant length (cm) at various DAP			
	15	30	45	60
P0	20.78	43.90 b	49.79 b	23.88
P1	20.58	37.85 a	43.46 a	29.33
P2	21.50	38.50 a	40.08 a	21.50
P3	21.64	36.85 a	41.90 a	27.12
P4	21.96	43.83 b	42.67 a	26.71
P5	21.10	42.10 ab	40.52 a	27.90
LSD 5%	NS	5.29	3.22	NS
CV	5.90	8.67	8.48	25.07

Notes: Numbers followed by the same letters for the same columns show no significant difference based on the LSD (Least Significant Difference) 5% test. CV= Coefficient of variance. DAP = Days after planting. NS = Non significant.

Table 3: Average Number of Leaves of Shallot with Various Weed Controls Treatments.

Treatments	Number of Leaves (leaves / plants) at Observation (DAP)			
	15	30	45	60
P0	11.98	13.83 a	14.08 a	3.96 a
P1	14.20	17.95 c	30.79 d	9.46 c
P2	12.40	14.30 ab	22.54 bc	6.10 ab
P3	13.00	14.95 ab	23.92 c	7.42 b
P4	12.95	14.79 ab	20.54 b	5.33 ab
P5	12.30	15.30 b	21.29 bc	5.60 ab
LSD 5%	ns	1.66	3.22	3.26
CV	8.22	7.24	9.63	34.25

Notes: Numbers followed by the same letters for the same columns show no significant difference based on the LSD (Least Significant Difference) 5% test. CV= Coefficient of variance. DAP = days after planting. NS = non significant.

Table 4. Average Number of Tillers of Shallots In Various Weed Control Treatments.

Treatments	Observed number of tillers (tillers/ plants) at various DAP			
	15	30	45	60
P0	3.21	3.83	4.25	3.33 a
P1	3.79	4.75	5.04	4.83 b
P2	3.58	4.20	4.25	4.80 b
P3	3.46	4.12	4.25	4.75 b
P4	3.67	4.13	4.25	4.00 b
P5	3.41	4.20	4.46	5.00 b
LSD 5%	NS	NS	NS	0.87
CV	10.62	8.27	10.52	12.95

Notes: Numbers followed by the same letters for the same columns show no significant difference based on the LSD (Least Significant Difference) 5% test. CV= Coefficient of variance. DAP = Days after planting. NS = Non significant.

Table 5: Average Fresh Weight of Shallot Bulbs In Various Weed Control Treatments.

Treatments	Observed fresh weight of bulbs (g/ plant) at various DAP			
	15	30	45	60
P0	1.06	3.46 a	6.96 a	11.46 a
P1	1.48	5.67 bc	23.13 d	41.21 c
P2	1.30	5.60 bc	17.88 bc	33.58 b
P3	1.26	5.21 b	19.85 cd	34.17 b
P4	1.35	6.38 c	15.38 bc	33.75 b
P5	1.40	5.10 b	13.63 b	32.13 b
LSD 5%	NS	1.02	5.44	4.56
CV	14.76	12.92	22.38	9.76

Notes: Numbers followed by the same letters for the same columns show no significant difference based on the LSD (Least Significant Difference) 5% test. CV= Coefficient of variance. DAP = Days after planting. NS = Non significant.

Tabel 6: Average Dry Weight of Shallot Bulbs In Various Weed Control Treatments..

Treatment	Observed dry weight of bulbs (g/ plant) at various DAP			
	15	30	45	60
P0	0.38	0.98 a	4.40 a	7.96 a
P1	0.45	1.41 b	15.71 c	32.12 c
P2	0.42	1.00 a	12.83 bc	26.30 b
P3	0.41	1.05 a	13.67 bc	26.08 b
P4	0.44	0.95 a	11.63 bc	26.58 b
P5	0.43	0.90 a	10.00 b	25.30 b
LSD 5%	NS	0.27	4.37	11.66
CV	24.48	17.10	25.50	4.25

Notes: Numbers followed by the same letters for the same columns show no significant difference based on the LSD (Least Significant Difference) 5% test. CV= Coefficient of variance. DAP = Days after planting. NS = Non significant.

At 30 DAP and 45 DAP, the length of plants with the P0 treatment (without weed control) was 43.90 cm and 49.79 cm, significantly longer than with the other treatments. Plant length with the the P1 treatment (weed free) was of 37.85 and 43.46 cm and not significantly different from the P2 (weeding at 15, 30 and 45 DAP), P3 (herbicide application + weeding at 30 DAP), P4 (silver black plastic mulch + weeding at 30 DAP) and P5 (rice straw mulch + weeding at 30 DAP treatments. For the number of leaves, there was no difference among treatments of weed control as observed at 15 DAP and there were significant differences as observed at 30, 45 and 60 DAP (Table 3). Observation made at 30,45 and 60 DAP showed that the number of leaves with the P1 (weed-free) treatment (17.95, 30.79, 9.46/ plant) was significantly higher than that of the other treatments. The number of leaves was significantly lower in the P0 treatment (without weed control) being 13.83, 14.08, 3.96/ plant. The number of leaves with P2 (weeding at 15, 30 and 45 DAP), P3 (herbicide application + weeding at 30 DAP, P4 (silver black plastic mulch + weeding at 30 DAP and P5 (rice straw mulch + weeding at 30 DAP) treatments were generally no different. Murthy, et al. (2009) reported that the number of leaves was significantly higher in weed-free treatment up to 60 days after planting.

In the number of tillers there was no difference between weed control treatments at observations of 15 DAP to 45 DAP (Table 4). At 60 days after planting, the number of tillers in weed-free treatment (4.83 tillers/ plant) did not differ from other weed control treatments (4.00 - 5.00 tillers/ plant). The number of tillers was significantly lower in the treatment without weed control (P0) being 3.33 tillers/ plant.

For the fresh weight of the bulbs there was no difference among weed control treatments as observed at 15 DAP (Table 5). At 30 DAP, the fresh weight of bulbs with the P1 (weed-free) treatment was 5.67 g/ plant and was not different from P4 (silver black plastic mulch + weeding at 30 DAP) treatment, being 6.38 g/ plant and P2 (weeding at 15 DAP, 30 DAP and 45 DAP) treatment, being 5.60 g/ plant. At 45 DAP , the fresh weight of bulbs with the P1 (weed-free) treatment was 23.13 g/ plant and was not significantly different from P3 (herbicide + weeding application at 30 DAP) treatment, which of 19.85 g/ plant. The fresh weights of bulbs with other weed control treatments were lower. Furthermore, at 60 days after planting, the fresh weight of bulbs was significantly

heavier at P1 (weed free) treatment, being 41.21 g/ plant. The fresh weight of bulbs was significantly heavier with P2 (weeding at 15 DAP, 30 DAP and 45 DAP), P3 (herbicide application + weeding at 30 DAP), P4 (silver black plastic mulch + weeding at 30 DAP) and P5 (rice straw mulch + weeding at 30 DAP) compared to P0 treatment (without weed control). The fresh weight of bulbs was significantly lower with the P0 treatment (without weed control) as observed at 15 DAP up to 60 DAP, weighing 1.06, 3.46, 6.96 and 11.46 g/ plant respectively.

The dry weights of bulbs/ plant among weed control treatments did not differ as observed at 15 DAP (Table 6). At observed at ages 30, 45 and 60 DAP, the dry weight of bulbs/ plant was significantly heavier with the P1 (weed-free) treatment, being 1.41, 15.71 and 32.12 g/ plant. The significantly lowest dry weight of bulbs was with P0 (without weed control) treatment, being of 0.98, 4.40 and 7.96 g/ plant. Meanwhile, the dry weight of bulbs with the P2 (weeding at 15 DAP, 30 DAP and 45 DAP), P3 (herbicide application + weeding at 30 DAP), P4 (silver black plastic mulch + weeding at 30 DAP) and P5 (rice straw mulch + weeding at 30 DAP) 30 DAP) treatments was significantly higher than with P0 (without weed control) treatment.

3.3 Yield Component

Weed control treatment significantly affected the fresh weight of bulbs/ plants, dry weight of bulbs/ plants, dry weight of bulbs/ harvest plot (0.47 m2) and crop yields/ ha (Table 7). The number of bulbs/ plant did not show a difference among weed control treatments. For the P1 (weed-free) treatment, the fresh weight of bulbs per plant (47.96 g/ plant), dry weight of bulbs/ plant (39.21 g/ plant), dry weight of bulbs/harvest plot (687.75 g/ 0.47 m^2) and yield/ha (12.08 t/ ha) were significantly higher than other weed control treatments, and significantly lowest for P0 (without weed control) treatment, being 3.05 t/ ha. The yield component for P2 (weeding at 15 DAP, 30 DAP and 45 DAP), P3 (herbicide application + weeding at 30 DAP), P4 (silver black plastic mulch + weeding at 30 DAP) and P5 (rice straw mulch + weeding at 30 DAP) treatments showed no difference and were significantly higher compared to P0 (without weed control) treatment. Weed index was significantly lower for the P2, P3, P4 and P5 treatment, being 13.12, 18.39, 17.99 and 19.12 compared to P0 of 74.77. Uygur, et al. (2010) stated that the highest onion yield was for weed-free treatment, followed by oxadiazon herbicide and oxyfluorfen herbicide. Poddar, et al. (2017), stated that the application of oxyfluorfen herbicide at a dose of 200 g/ ha + weeding at 30 days significantly decreased the weed density and dry weight and increased the bulb yield of onion. Qosem (2015) reports that onion yields decrease by 87% if weeds are not controlled during the growth period of the plant.

Table 7: Average Yield Components of Shallots on Various Weed Control Treatments.

Treatments	Average					
	Number of Bulbs (bulbs / plants)	Bulbs Fresh Weight of (g / plant)	Bulbs Dry Weight (g/ lant)	Bulbs Dry Weight (g/ harvest plot)	Yield (t/ha)	Weed Index
P0	5.25	13.96 a	10.63 a	173.50 a	3.05 a	74.77
P1	5.58	47.96 c	39.21 c	687.75 c	12.08 c	-
P2	5.20	40.40 b	33.50 b	597.50 b	10.50 b	13.12
P3	5.25	39.83 b	33.17 b	561.25 b	9.86 b	18.39
P4	5.71	40.21 b	33.58 b	564.00 b	9.91 b	17.99
P5	5.20	36.80 b	29.90 b	556.30 b	9.77 b	19.12
LSD 5%	ns	5.45	4.97	74.22	0.62	
CV	9.55	9.91	11.00	9.42	9.42	

Notes: Numbers followed by the same letters for the same columns show no significant difference based on the LSD (Least Significant Difference) 5% test. CV= Coefficient of variance. DAP = Days after planting. NS = Non significant.

IV. CONCLUSION

Weed control has a significant effect in controlling weeds and increasing the growth and yield of shallots. With the weed-free treatment, weed dry weight was significantly lower,while plant growth and yield /ha significantly increased. The P2 (weeding at 15 DAP, 30 DAP and 45 DAP), P3 (application of oxyfluorfen herbicide with a dose of 1.5 l /ha + weeding at 30 DAP), P4 (silver black plastic mulch + weeding at 30 DAP) and P5 (rice straw

mulch + weeding at 30 DAP) treatments had significant effects in controlling weed growth as well as increasing the growth and yield of shallots compared to without weed control.

ACKNOWLEDGEMENTS

This research was funded by Research Grant for Professor from the Rector of Brawijaya University and Agricultutre Faculty, Brawijaya University (PNBP.DIPA-042.01.2.400919/2019).

REFERENCES

[1] Center for Data and Agriculture and Information System. 2016. Outlook of Onion. Center for Data and Agriculture and Information System, Ministry of Agriculture. http://epubliksi.setjen.pertanian.go.id/file/301--outlook-bawang merah-2016.

[2] Bhullar,M.S., Kaur,T., Kaur, S. and Yadav, R. 2015. Weed management in vegetable and flower crop-based system. Indian Journal of Weed Science 47(3): 277-287. http://isws.org.in/IJWSn/File/2015_47_Issue-3_277-287.pdf.

[3] Kumar, U. 2014. Weed management studies in onion (Allium cepa L.). The Asian Journal of Horticultura 9(2): 426-430. DOI:10.15740/HAS/TAJH/9.2/426-430.

[4] Murthy, K. N.K., Fathima,P.S. and Vidya, A. 2009. Effect of crop weed competition on the performance of direct seeded onion (Allium cepa L.). International Journal of Agricultural Science 5(2): 558- 583. https://pdfs.semanticscholar.org/7bf4/d3aa4eb05d466bc75dc0724dc95d28013e74.pdf.

[5] Poddar, R., Bera, S. and Ghost, R.K. 2017. Weed management in onion through oxyfluorfen and its effect on soil microflora and succeeding crop of balckgram. Indian Journal of weed science 49(1): 47-50. DOI:10.5958/0974-8164.2017.00012.0

[6] Prachand,S., Kubde, K.J. and Bankar,S. 2014. Effect of chemical weed control on weed parameters,growth, yield attributes,yield and economics in soybean (Glycine max). American Eurasian J.Agric.& Environ.Sci. 14(8): 698-701. DOI: 10.5829/idosi.aejaes.2014.14.08.12376.

[7] Priya,R.S., Chinnusamy,C., Janaki,P. and Arthanari.P.M. 2017. Persistence and carryover effect of oxyfluorfen residues in red sandy clay loam soil. Journal of Pharmacognosy and Phytochemistry 6(3): 527-532. http://www.phytojournal.com/archives/2017/vol6issue3/PartI/6-3-108-760.pdf.

[8] Qasem,J.R. 2005. Critical period of weed competition in onion (Alium cepa L.) in Jordan. Jordan Journal of Agriculture Science 1(1). https://journals.ju.edu.jo/JJAS/article/view/1286/5935.

[9] Sekara, A., Pokluda, R.,Vacchio,L.D., Somma, S., Caruso, G. 2017. Interaction among genotype,environment and agronomic practices on production and quality of storage onion (Allium cepa L.) – A review. Hort.Sci. (Prague) 44(1): 21-42. DOI:10.17221/92/2015-HORTSCI.

[10] Tripathy, P., Sahoo,B.B., Patel, D and Dash, D.K. 2013. Weed management studies in onion (Allium cepa L.). Journal of Crop and Weed 9(2): 210-212. DOI:10.13140/RG.2.2.27442.71360.

[11] Uygur, S., Gürbüz, R. and Uygur, F.N. 2010. Weed of onion fields and effects on some herbicides on weeds in Cukurova region, Turkey. African Journal of Biotechnology 9(42): 7037-7042. DOI.10.5897/AJB10.1005.

[12] Vijayvergiya, D., Ali, S.A., Das, M.P., Ramgiry, P. and Uikey,S. 2018. Effect of pre-emergence herbicides on weed control of kharif onion (Allium cepa L.) in vindhyan plateu of Madhya Pradesh. The Pharma Innovation Journal 7(1): 376-378. http://www.thepharmajournal.com/archives/2018/vol7issue1/PartF/7-1-42-359.pdf.

https://dx.doi.org/10.22161/ijeab.51.22 *ISSN: 2456-1878*

Productivity Optimization in Rice-Based Intercropping Systems of Central Uganda

Immaculate Mugisa[1], Benard Fungo[2], Stella Kabiri[1], Godfrey Sseruwu[1], Ruth Kabanyoro[1]

[1]Mukono Zonal Agricultural Research and Development Institute, National Agricultural Research Organization (NARO), P.O. Box. 164, Mukono, Uganda
[2]National Forestry Resources Research Institute, National Agricultural Research Organization (NARO), P.O. Box. 1752, Mukono, Uganda

Abstract*— *Upland rice production in Central Uganda is mainly done by small scale farmers for both food security and income generation. However, they are faced with a number of challenges including drudgery, birds that eat the crop, erratic weather and limited land holdings. Due to the inadequate land available to them, upland rice has to compete with other food crops for land for cultivation. Thus, apart from the conventional mono-crop, alternative cropping systems that enable them to grow rice while simultaneously benefiting from other major food crops are quite desirable to them.

This study was conducted to identify suitable upland rice-based intercropping alternatives to enable upland rice farmers to benefit from intercropping. Three experiments were conducted for two consecutive seasons on rice-beans, rice-groundnuts and rice-maize intercrops, each as a randomized complete block design with 5 treatments and 3 replicates. Treatments for the rice-beans experiment included sole rice, sole beans, intercrop 1(rice:beans in ratio 3:2), intercrop 2 (rice:beans in ratio 4:2) and intercrop 3 (rice:beans in ratio 4:3). The same was done for experiments on the other two intercrops. Data were collected on plant height, tiller number, grain yield of rice and yield of the three intercrops at harvest. Results indicated that intercropping rice with the three crops leads to more yield benefits as observed from the land equivalent ratios (LERs) obtained (average 1.5).The best intercrop with better yields and higher LERs was intercrop 3 for the rice-legume mixtures and rice-based intercrop 1 for the rice-maize mixture.

Keywords*— *Rice, Beans, Groundnut, Maize, Intercrop.

I. INTRODUCTION

Rice is one of the crops that has the potential to improve farmers' incomes and livelihoods (JICA, 2009), thereby contributing to socio-economic growth of many rural farm households in Uganda. It is an important food security factor (UNRDS, 2009) and staple crop in many rural and urban households throughout the country (Oryokot *et al*., 2004). Rice is now widely grown in many parts of Uganda, particularly in the eastern and northern regions (Kijima *et al*., 2011). In the past, rice production in Uganda was mainly limited to irrigation schemes that had been established by the government in the 1960's and 1970's. However, with the introduction of upland rice in the early 2000's, the trend is changing as more small scale and a few large scale farmers take on the relatively new enterprise (UNRDS, 2009).

The production of upland rice in Uganda is still to a large extent done by resource poor farmers who hardly use external inputs. Therefore, the yields and consequently incomes from upland rice growing are still low compared to paddy rice (Defoer *et al*., 2004). In Central Uganda, the production of upland rice by small scale farmers in particular has been linked with challenges including erratic weather and droughts, birds that eat the crop at maturity, drudgery (particularly at planting, weeding and bird scaring) and theirlimited land holdings, where upland rice competes with food crops for land for cultivation. Most upland rice farmers are accustomed to monocultures as they've been trained, but they think that rice takes up land for long, depriving them of land for growing other important food crops like beans, maize, bananas and cassava (MUZARDI, 2013). Thus, they would wish to have an alternative cropping system that enables them to grow rice while at the same time benefiting from their usual food crops. The present system of mono-cropping has failed to meet the diversified domestic needs of small holder farmers from the

https://dx.doi.org/10.22161/ijeab.51.22 *ISSN: 2456-1878*

dwindling supply of new lands for cultivation and other limited resources (Farrukh *et al*., 2000). These conditions necessitate a shift from mono-cropping to intercropping, which is considered as an excellent strategy for intensifying land use, absorbing excess labour and increasing income and production per unit area and time (Willey, 1979). Intercropping refers to the growing of two or more crops simultaneously on the same piece of land in alternate rows or set of rows (Zandstra, 1979). In India, rice intercropping practices have for a longtime been carried out. According to Mandal *et al*, 1990, intercropping provides farmers with profit and subsistence-oriented requirements from the same piece of land. This conclusion was drawn after a two-year experiment on intercropping rice and legumes to assess the effect of legumes on rice yields. Elsewhere in Nigeria, a study conducted on the rice-cowpea intercrop by Oroka and Omoregie (2007) revealed that nitrogen use efficiency is higher in intercrops than sole crops. In Nepal, a study was carried out to improve soil fertility and enhance productivity of upland rice varieties (Rokaya, 2004). In this study, upland rice varieties and legumes were grown in intercrops. Investigations indicated that continuous inclusions of legumes have positive impact on soil fertility, resulting in the sustainable productivity of upland rice.

Beyond its importance as a farming practice, intercropping often, offers the possibility of yield advantages relative to sole cropping through yield stability and improved yield (Willey, 1979). Contributors to yield advantages include; better use of growth resources (Trenbath, 1986) and better control of weeds, pests and diseases (Willey, 1979). It also helps to maintain soil fertility (Patra *et al*., 1986), making efficient use of nutrients (Ahmed & Saeed, 1998) and ensuring economic utilization of land, labour and capital resources (Singh et al., 1996). Intercropping may be practiced for a number of yield goals, not limited to the production of dry matter (Willey, 1979). Morris and Garity (1993), for instance, stated that water use efficiency in intercropping was 18.99% higher than that in sole cropping. In a study that was conducted in Central Uganda in 2008 on factors affecting rice production in Semuto, Nakaseke District, 30.7% of the 150 rice farmers surveyed were found to be haphazardly intercropping rice with beans and maize despite their having been sensitized on rice growing as a mono-crop (MUZARDI, Unpublished).Farmers revealed that they were carrying out this practice in order to increase on their income, food security and to maximize the utilization of their small land holdings.

Since the conventional method of planting rice in monocultures does not permit intercropping, a study was initiated to identify suitable upland rice-based intercropping systems to ensure that farmers maximize the benefits of intercropping. Introducing other crops into the upland rice intercropping system would provide an opportunity for resource poor farmers to get cash from rice without entirely compromising on their food security. Moreover, in light of the current climatic changes and unreliable rainfall patterns, an upland rice farmer would not entirely make losses in case of crop failure due to unanticipated droughts but would still benefit from the secondary crop.

II. MATERIALS AND METHODS

2.1 Study Area

The study was carried out in Mukono (coordinates: $00^0$20'N, $32^0$45'E), Kayunga (01^0 00′N, 32^0 52′E) and Kiboga ($01^0$00'N,$31^0$46'E) districts, all located within the Lake Victoria Crescent Agro-ecological Zone of Uganda.

2.2 Experimental design

Three experiments were conducted in 2013on selected rice-based intercrops: rice-beans, rice-maize and rice-groundnuts, for two consecutive planting seasons in 2013, each as a randomized complete block design (RCBD) with five treatments and 3 replicates. Treatments for the rice-beans experiment included sole rice, sole beans, intercrop 1(rice: beans, 3:2 rows), intercrop 2 (rice: beans, 4:2 rows) and intercrop 3 (rice: beans, 4:3 rows). The same was done for experiments on the other two rice-based intercrops: rice-maize and rice-groundnuts. The row-intercropping method was used and both rice and each of the intercrops were planted at the same time in different proportions in alternate rows (Fig.1).

https://dx.doi.org/10.22161/ijeab.51.22 ISSN: 2456-1878

Fig.1: A trial field in Mukono district planted with the rice-beans 3:2 row intercrop

Upland rice variety NERICA 4 was used as the base (main) crop. A plot size of 8m X 5m was maintained for each experimental unit. The rice seed was directly planted using the drilling method leaving a spacing of 30cm between rows. All intercrops (beans, maize and groundnuts) were directly seeded with one seed per hill at a spacing of 45cm x 15cmfor groundnuts and beans and 75cm x 50cm for maize. The wider spacing of the intercrop was always maintained between rows that bordered the rice. Prior to laying out the experiments, soil samples were taken from the trial sites for laboratory analysis to determine their baseline fertility status. The soil properties analyzed included organic matter content, pH, total nitrogen (N), phosphorus (P) and selected base cations (Ca, Mg and K). Fertilizer application was then done using DAP (at 20 days after emergence (DAE)) and Urea (at 20 and 60 DAE). The trials were weeded twice during each season and were entirely rain-fed- no irrigation was done.

2.3 Data collection and Analyses

Data was collected on plant height, tiller number and grain yield of the rice crop. Data was also taken on yields of the intercrops i.e. beans, maize and groundnuts. At crop maturity, yields were obtained by harvesting the different crops within each net plot and extrapolating yields to kg/ha. This data was analyzed using GenStat statistical package (Genstat, 2010) to generate analyses of variance (ANOVA) to compare the yield differences of the different treatments.

The Land Equivalent Ration (LER), defined as the relative land area required as a sole crop to produce the same yields as intercrops, was obtained by using the following formula (Mead and Willey, 1980).

$$LER = Pr + Pb = \left[\frac{Yr}{Sr}\right] + \left[\frac{Yb}{Sb}\right]$$

Where:

Pr and *Pb*= partial LER (rice and beans respectively)

Yr and *Yb*= intercrop yields (rice and beans respectively)

Sr and *Sb*= yields of sole crops (rice and beans respectively).

An LER value of 1, signifies no advantage in intercropping as compared to sole cropping, whereas LER > 1 means a larger area of land is needed to produce the same yield of both sole crops of each component, in relation to intercropped mixture. An LER < 1.0 shows a disadvantage of intercropping in comparison to mono cropping (Oroka and Omoregie, 2007; Kutrata, 1986).

One-way Analysis of Variance (ANOVA) was also used to test the difference in the mean LER among the treatments (at *P = 0.05).* Descriptive statistics were used to characterize the different sites in terms of soil properties.

III. RESULTS AND DISCUSSION

3.1 Yields and Land Equivalent Ratios

Overall, analysis of variance of trial results revealed that there were no significant differences ($p \leq 0.05$) between the yields of rice planted in the different treatments (Table 1). However, there were significant differences ($p \leq 0.05$) in the yields of the

https://dx.doi.org/10.22161/ijeab.51.22 *ISSN: 2456-1878*

various treatments for all the intercrops (maize, beans and groundnuts) (Table 2). The interaction between site, season and treatment was also significant ($p \leq 0.05$). This implies that the yield of rice was more or less similar in the various rice-based intercrops studied whereas the intercrop yields varied to a larger extent when intercropped with rice in different proportions. This could be attributed to the higher plant densities of rice compared to the intercrops considering the row intercrop ratios that were used in this study which all had more rows of the rice crop. A previous study carried out on a rice-cowpea intercrop by Oroka and Omoregie (2007) revealed that there was a response of rice yield to intercrop density such that higher densities of rice in cereal-legume intercrops do not substantially decrease its grain yield in comparison to intercrops with lower rice densities.

Table 1: Summary of ANOVA results for the yields of rice intercropped with three crops

Source of Variation	d.f	F-value		
		Rice-maize	Rice-beans	Rice-groundnuts
Season	1	<0.001*	0.006*	<0.001*
Site	2	<0.001*	0.723	0.011*
Treatment	4	0.066	0.233	0.668
Season*Site	2	0.279	0.594	0.024*
Season*Treatment	4	0.898	0.638	0.878
Site*Treatment	8	0.919	0.366	0.987
Season*Site*Treatment	8	0.529	0.214	0.450
Residual	71			

*Indicates significance at $p \leq 0.05$

Table2: Summary of ANOVA results for the yields of intercrops

Source of Variation	d.f	F-value		
		Maize	Beans	Groundnuts
Season	1	0.246	0.014*	<0.001*
Site	2	0.371	0.481	0.004*
Treatment	4	<0.001*	<0.001*	<0.001*
Season*Site	2	0.117	<0.001*	<0.001*
Season*Treatment	4	0.384	0.957	0.065
Site*Treatment	8	0.798	0.896	0.359
Season*Site*Treatment	8	0.659	0.011*	<0.001*
Residual	71			

*Indicates significance at $p \leq 0.05$

3.2 Rice yields, Intercrop yields and Land Equivalent Ratios

Yield results revealed that intercrop 1 (3:2 row intercrop), produced the highest yields of rice (Table 3), followed by the treatment of sole rice. It also produced the 2nd highest number of tillers after the rice sole crop. However, it had the lowest plant height compared to all other treatments. The 2nd best performing treatment in terms of average rice yields was intercrop 3 (4:3 row intercrop).

https://dx.doi.org/10.22161/ijeab.51.22 *ISSN: 2456-1878*

Table 3: Summary of grand means of selected rice parameters in the four treatments

Treatment (plant proportions; Rice: intercrop)	Yields (kg/ha)	Plant height (cm)	Tiller No.
Intercrop 1 (3:2)	2,560	34.6	10.12
Intercrop 2 (4:2)	2,058	38.7	9.88
Intercrop 3 (4:3)	2,134	39.8	10.07
Sole rice	2,357	36.2	11.3

In the rice: maize trial, intercrop 1 (3:2 row intercrop) produced the highest maize yields, followed by intercrop 3 and 2 respectively. For the case of beans and groundnuts, intercrop 3 produced the highest yields followed by intercrops 1 and 2 respectively (Fig.2). These results reveal that intercrop 3 (4:3 row intercrop) is the best performing in terms of rice yields for row intercropping of rice with beans and groundnuts whereas intercrop 1 produces higher rice yields for the case of rice: maize row intercropping. The generally low average yields obtained for both rice and the intercrops could be due to the low precipitation that occurred in the study areas during the 2nd planting season of the study since the trials were rain-fed and not irrigated. Furthermore, it could also be due to the very low values of nitrogen (below critical values- as revealed by the soil analysis results in Table 4) at the trial sites despite the urea that was applied during the course of the study.

Results of LER revealed that overall, intercropping rice with beans, ground nuts or maize, yields more than each of the crops grown alone (Fig.3), implying an advantage of intercropping over mono-cropping, thus better resource use efficiency by the crop mixture.

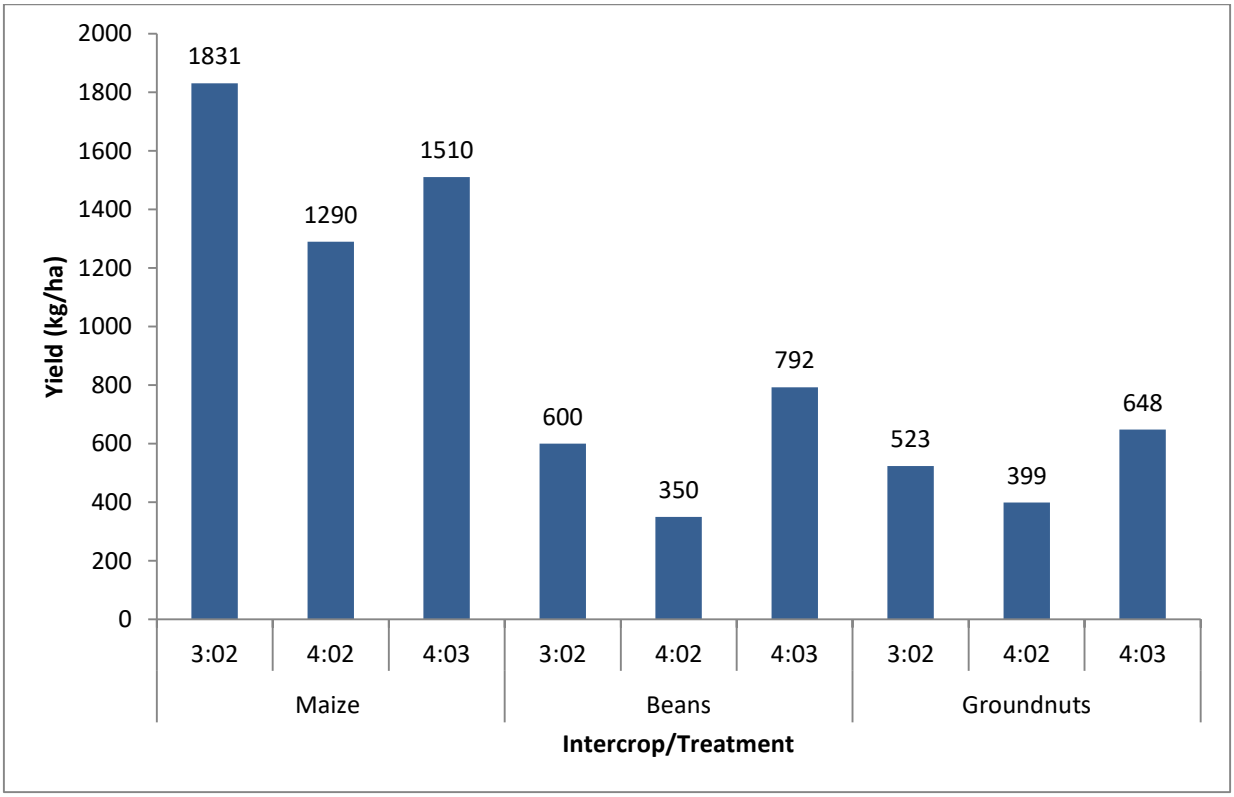


Fig.2: Mean yields of intercrops in treatments (kg/ha)

https://dx.doi.org/10.22161/ijeab.51.22 *ISSN: 2456-1878*

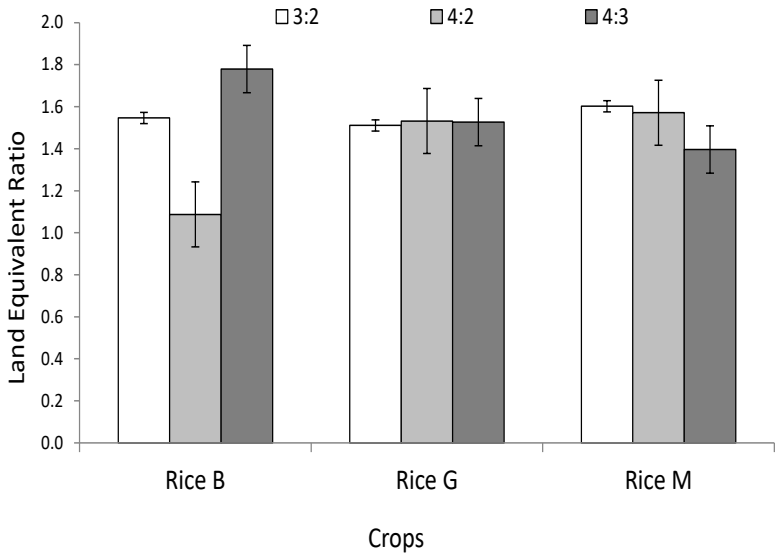


Fig.3: Overall LERs of intercrops

Rice B, Rice G and Rice M represent Rice intercropped with Beans, Groundnuts and Maize respectively. R:X indicates number of rice: intercrop rows, respectively

LER measures the levels of intercrop interference going on in the cropping system. A total LER higher than 1.0 indicates that the presence of positive inter-specific interference that exists in the mixture is not as intensive as the inter-specific interference that exists in the monoculture (Dariush, *et al*., 2006). An LER value of 1.0 indicates no difference in yield between the intercrop and the monocrop and any value greater than 1.0 indicates an advantage for the intercrop. An LER of 1.2, for instance, indicates that the area planted with the mono-crop would need to be 20% greater than the area planted with intercrop for the two to produce the combined yield (Kutrata, 1986).The average value of LER obtained (1.5) across intercrops in both seasons indicates that on average approximately 1.5 times more land is required to produce the same yield of the sole crops compared to that of the intercrops. Values of LER >1 were obtained in another study on rice based intercrops in Nigeria by Oroka and Omoregie (2007) where a yield advantage of intercropping at all levels of nitrogen and plant densities were reported for a rice-cowpea mixture with yield advantages ranging from LER 1.79 to 2.30. The rice-beans and rice-groundnuts intercrop, both produced better yields and higher LERs under intercrop 3 (4:3 row intercrop) which had higher intercrop densities compared to intercrop 2 (4:2 row intercrop) and intercrop 1 (3:2 row intercrop). This yield advantage could be attributed to more efficient utilization of light, water and nutrients during the growing season considering that the legumes are much shorter than the rice. Beans in particular have a shorter life cycle compared to rice, which must further have contributed to its best performance in terms of LER. Differences in growth cycles between crops has been reported to be important in intercropping because they enable more efficient water and nutrient utilization during the growth period (Willey and Osiru, 1972).

In a rice-cassava intercrop study that was done in Sierra Leone (Dahniya *et al*., 1994), it was reported that a higher intercrop density leads to lower values of LER and vice versa. For the case of this study, this was only observed in the rice- maize trial where higher densities of maize in intercrop 3 (4:3 row intercrop) led to a lower LER whereas the lowest maize density intercrop gave the highest LER. This could also be attributed to the less efficient light, water and nutrient utilization by the two crops which are both tall and take about the same time to mature in the field since these trends were the opposite for the rice-beans and rice-groundnuts intercrop.

3.3 Soil Properties of study sites

https://dx.doi.org/10.22161/ijeab.51.22 *ISSN: 2456-1878*

Table 4: Soil properties of the three experimental sites

Site	Statistics	pH	*OM (%)	Total N (%)	P (ppm)	K (ppm)	Ca (ppm)	Mg (ppm)
Kayunga	**Mean**	**6.65**	**3.21**	**0.18**	**8.4**	**447**	**4185**	**489**
	SD	0.62	0.68	0.03	11.6	205	1398	141
	N	58	58	58	58	57	58	58
Kiboga	**Mean**	**6.23**	**5.47**	**0.25**	**15.8**	**404**	**3378**	**695**
	SD	0.75	1.45	0.05	10.5	161	1292	296
	N	45	45	45	45	45	45	45
Mukono	**Mean**	**5.98**	**4.86**	**0.18**	**24.3**	**382**	**2826**	**632**
	SD	0.56	1.07	0.04	28.2	228	1054	152
	N	45	45	45	45	45	45	45
Sufficient levels		**5.2-7.0**	**6.0**	**0.3**	**20.0**	**2000.0**	**5**	**600.0**
Critical values		**5.2**	**3.0**	**0.2**	**5.0**	**350.0**	**150.0**	**100.0**

** OM= Organic Matter*

Soil pH of all the sites was within the optimum range required for most crops. Organic matter (OM) content was above critical limits but also below sufficient level for all the sites. The macro-nutrients nitrogen, phosphorus and potassium were at or below critical limits for all the sites. That notwithstanding, the macro-nutrients Ca and Mg were not limiting.

The low level of OM reflects the combination of high degradability associated with high temperature conditions prevalent in the tropical region, as well as the relatively low level of OM inputs by farmers. This is due to the tendency for farmers to remove biomass residues from crop fields during harvesting without replacing it. The biomass is often used as either feedstock or burnt during land clearing for subsequent seasons. The major macro-nutrients (N, P and K) are those usually consumed in large quantities and can easily be replaced through commercial synthetic fertilizers. The exceeding low level of nitrogen at the trial sites could have contributed to the low yields despite the blanket application of urea during the course of the study.

IV. CONCLUSIONS AND RECOMMENDATIONS

The results of the present study revealed that intercropping rice with beans, ground nuts or maize, yields more than each of the crops grown alone as shown by the LER values obtained. On average approximately 1.5 times more land is required to produce the same yield of the sole crops compared to that of the intercrops. Furthermore, it was observed that the yield of rice was more or less similar in the various rice-based intercrops studied whereas the intercrop yields varied to a larger extent when intercropped with rice in different proportions. It can be concluded therefore that using higher plant densities of rice in comparison to the intercrop will more or less not affect the rice yields obtained. We therefore recommend that small holder farmers who are interested in rice intercropping utilize higher ratios of rice compared to intercrop using the row intercropping method.

An analysis of the three rice- based intercrops studied showed that the best row intercrop the rice-beans and rice-groundnuts mixtures was intercrop 3 which had a planting ratio of 4:3 (rice: intercrop) and produced both better yields and higher LERs. However, for the case of rice-maize, intercrop 1 with a planting ratio of 3:2 (rice: maize) performed best. We therefore recommend the 4:3 row intercrop ratio for rice-based intercrops with beans and groundnuts whereas 3:2 is preferable for rice-maize intercrops. This would ensure enhanced yields and ensure food security among smallholder upland rice farmers as a result of improved nutrient, light and water use efficiency in these intercrops.

Soil productivity in rice based intercrops is mainly limited by low nitrogen, phosphorus, potassium and organic matter. In order to improve soil productivity, the application of mineral fertilizer such as NPK is recommended. Farmers are also advised to retain crop residues and/or employ biomass transfer systems (manure, plant residues from their places into the gardens) in order to increase the level of soil organic matter.

Through. Finally, extension education on appropriate row intercropping and fertility management practices would also help optimize nutrient use efficiencies from fertilizer use in the recommended intercrops.

ACKNOWLEDGEMENTS

This work was funded by the World Bank under the Agricultural Technology and Agribusiness Advisory Service (ATAAS) project implemented by the National Agricultural Research Organization. We are very grateful for this support. We appreciate all staff of MUZARDI who were involved in data collection including George Kituuka, Robinah Gafabusa, Barbara Kyampeire, Maria Nampeera, Robert Mutyabaas well as staff of Kiboga and Kayunga District Local Governments for their collaboration during the entire course of this study.

REFERENCES

[1] Ahmad, N. and Saeed, M. 1998. Resource-use efficiency of some wheat based intercropping systems at different patterns of wheat plantation. Pakistan Journal of Agricultural Science, 35:52-4

[2] Dahniya, M.T., Jalloh, A. and Ezumah, H.C. 1994. Intercrop yields of cassava and rice as influenced by cassava variety and population. In: Ofori, F.; Hahn, S.K. (Ed.). Root crops in a developing economy. Ibadan: International Institute for Tropical Agriculture, p.194-199.

[3] Dariush, M., Ahad, M. and Meysam, O. 2006. Assessing the land equivalent ratio (LER) of two corn (Zea mays L.) varieties intercropping at various Nitrogen Levels in Karaj, Iran. Journal of Central European Agriculture. 7(2) 359- 364.

[4] Defoer, T., Wopeires, M.C.S., Jones, M.P., Lanson, F., Erenstein, O. and Guei, G.R. 2004. Challenges and technical opportunities for rice based production systems for food security and poverty alleviation in sub-saharan Africa. FAO Rice Conference.

[5] Farrukh, S.M., Shamshad, H., Asghar, M. and Kashif, M. 2000. Bio-Economics of different upland rice-based intercropping systems under strip plantation. International journal of agriculture and biology. Vol 2., No. 4: pp: 294-296.

[6] Genstat, 2010. Genstat Thirteenth Edition, Procedure Library Release PL21.3. Lawes Agricultural Trust, Rothamsted Experimental Station. Clarendon Press, Oxford.

[7] Jalloh, A., Dahniya, M.T. and Ezumah, H.C. 1994. Production of cassava-rice intercrop as influenced by cassava variety and time of introducing rice into cassava.Acta Horticulturae. 380, 200-204. DOI: 10.17660/ActaHortic.1994.380.30

[8] JICA. 2006. Japan International Cooperation Agency, (JICA) In collaboration with Sasakawa Africa Association Uganda. Final Survey Report on the status of rice production, processing and marketing in Uganda. Available on: https://www.mofa.go.jp/mofaj/gaiko/oda/bunya/agriculture/pdf/uganda_report.pdf (Accessed: 21st Dec. 2018)

[9] Kijima, Y., Otsuka, K. and Sserunkuuma, D. 2011. An inquiry into constraints of a green revolution in Sub-Saharan Africa: The Case of NERICA rice in Uganda. World Development. Vol. 39, No. 1, Pp. 77–86.

[10] Kutrata, T. 1986. A study on farming systems in USSA. Quarterly. J. Agro. Eco. 26: 179- 205.

[11] Mead, R. and Willey, R. W. 1980. The concept of land equivalent ratio and advantages. Experimental Agriculture. 16: 217- 226.

[12] Morris, R.A and Garrity, D.P. 1993. Resource capture and utilization in intercropping: non-nitrogen nutrients. Field Crop Research. 34:319-34.

[13] MUZARDI. 2013. Mukono Zonal Agricultural Research and Development Institute. Annual Report. 2013.

[14] Oryokot, J., Forster, M., Kayayo, B. and Pookat V. 2004. Private/Public sector partnerships in sustainable development and promotion of rice for improved farm household income in Uganda. Unpublished Paper Presented at the "Year of Rice Conference", Uganda.

[15] Patra, D.P.,Sachdev,M.S. and Subbiah, B.V. 1986. Nitrogen fixation by tropical elements. Fertil.Soils, 2: 165-71.

[16] Rokaya, B. B. 2007. Productivity improvement in upland rice through the inclusion of legumes, in far-western mid-hill areas of Nepal. 5th International Crop Sci. Congress, 10-15 September 2007, Adelaide, South Africa.

[17] Singh, G., Singh, O.P., Kumar, M., Rajput,A.L. and Maurya, S. 1996. Effect of intercrops on yields and economics, weeds and pest infestation of deep water rice. Annals of Agricultural Research. 17:14-17.

[18] Trenbath, B.R. 1986. Resource use by intercrops. In. Multiple cropping systems. C.A. Francis. MacMillan Pub. Co., New York, pp: 57-81.

[19] UNRDS. 2009. Uganda National Rice Development Strategy. 2nd Draft. Ministry of Agriculture Animal Industry and Fisheries. Available from: https://www.jica.go.jp/english/our_work/thematic_issues/agricultural/pdf/uganda_en.pdfAccessed: 21st Dec. 2018)

[20] Willey, R.W. 1979. Intercropping- its importance and research needs. 1. competition and yield advantages. Field crop Absts., 32: 1-10.

[21] Willey, R.W. and Osiru, D.S.O. 1972. Studies on mixture of maize and beans (*Phaseolus vulgaris*) with particular reference to plant population. Journal of Agricultural Science, v.77, p.531- 540.

[22] Zandstra, H. G. 1979. Terminologies in cropping systems "Definitions and Parameters"- Lecture notes of six month cropping systems training programme at IRRI, Philippines.

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

Selling Points of Sewage Sludge as an Enhancing Agent of Bioremediation of Diesel Oil-Polluted Soil

Kingsley Tochukwu Ughamba[1,2*], Nnabueze Darlington Nnaji[2], Kenneth Ejike Ogbonna[1,3], Chukwudi Anyanwu[2]

[1]Department of Science Laboratory Technology, University of Nigeria, Nsukka, Nigeria
[2]Department of Microbiology, University of Nigeria, Nsukka, Nigeria
[3]Department of Biochemistry, University of Nigeria, Nsukka, Nigeria
*Corresponding author: E-mail: kingsley.ughamba@unn.edu.ng

***Abstract*— *Bioremediation employing the action of microbes alone has been shown to be inadequate. The aim of this study was to evaluate the efficacy of sewage sludge (SS) in enhancing bioremediation of diesel oil-polluted soil. Diesel oil was introduced into the soil at the concentration of 10 % (v/w) and mixed with 5%, 10% and 15% (w/w) of sewage sludge. The remediation of the oil was determined gravimetrically using n-hexane as extractant. Effectiveness of the remediation strategy was assessed by the seed germination toxicity test. At the end of forty-two days, 32.22 % oil loss was recorded in the unamended polluted soil while 58.33% oil loss was recorded in the soil amended with sewage sludge. Hydrocarbon- utilizing bacteria (HUB) counts were significantly higher ($P \leq 0.05$) in the sewage sludge-amended options, ranging from $5.3 \pm 0.9 \times 10^6$ to $12.3 \pm 0.75 \times 10^6$CFU/g soil, as compared to the unamended control soil which gave 1.0×10^6- 3.8×10^6CFU/g of soil. The hydrocarbon-utilizing bacteria isolated from both the control and amended soils were identified tentatively as Bacillus cereus, Pseudomonas putida, Micrococcus varians, Corynebacterium sp, Acinetobacter sp and Bacillus licheniformis based on their cultural, morphological and biochemical characteristics. The fungal counts in the SS-amendment options were also higher than was recorded in the control option ranging from $3.8 \times 10^5 \pm 0.2$ to $11.6 \times 10^5 \pm 0.25$.Aerobic fungi isolated were Aspergillus niger, Aspergillus flavus, Fusarium sp, Cladosporium sp and Penicillium sp. The highest oil loss and germination indices were recorded in SS-amended options. There was a significant difference ($P \leq 0.05$) in oil loss and germination index between the unamended control soil and amended soil.*

Keywords*— *Bacteria, sewage sludge, germination, diesel oil, pollution.

I. INTRODUCTION

Petroleum-based products are the major energy source for vehicles, daily life and industry owing to their high energy content. Exploration, mining and transport of the same in developing countries have led to serious environmental hazards due to accidental spills [1]. The increasing use of diesel in car engines, industrial trucks and generators has led to a marked increase in the demand for diesel fuel in Nigeria and damages due to soil contamination by diesel oil may be extensive and have long term effect [2]. Release of hydrocarbons into the environment, whether accidentally or due to anthropogenic activities, is a main cause of water and soil pollution.

Diesel oil is a mixture of aromatic compounds and alkanes and is usually documented as soil pollutants because of their frequent release from accidental spills and leakage from storage tanks [3].Diesel is a medium-weight petroleum fuel with a boiling point range of 175^0C to 355 ^{0}C [4]. It is composed of over 200 petroleum hydrocarbon compounds corresponding to the molecular weight range of C_{10}-C_{28} alkanes [5,6].The exhaust fumes of diesel contains up to forty air pollutants including many suspected or known carcinogenic substances such as arsenic, formaldehyde and benzene. It also contains other harmful environmental pollutants, including nitrogen oxide, currently the single most important ozone-depleting emission [7].

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

Oil exploration and exploitation are very lucrative and major revenue earner in Nigeria. However, like most industrial activities, it produces environmental hazards that are "slow poisons" in that they often take months and years to cause disease and death. The slow and obvious environmental hazards occasioned by exploration of oil and exploitation of the same make it difficult to fully understand their impart in the health of Nigeria as a people, especially in the oil-bearing communities, even with the emergence of non-communicable diseases as major causes of ill health in Nigeria.

The need for remediating diesel oil-polluted areas has induced development of new technologies to detoxify contaminants not only through chemical or physical methods which are expensive, but through biological techniques as well. Bioremediation is an eco-friendly and cost-effective option that removes contaminants or renders them innocuous using natural biological activity. Microorganisms degrade these compounds by using enzymes in their systems and can be useful in cleaning up contaminated sites [8]. However, bioremediation employing the action of microbes alone has proven rather inadequate. Lack of essential nutrients such as nitrogen has been identified as a major limiting factor in petroleum hydrocarbon degradation among various factors. Therefore, the addition of organic or inorganic nutrients rich in nitrogen content remains an effective approach to enhance bioremediation process [9]. The use of organic wastes to stimulate the indigenous hydrocarbon degraders have been widely demonstrated [10, 11, 12]. However, few works have been done on the use of sewage sludge in enhancing the bioremediation of diesel oil-impacted soils. The present study was therefore undertaken to assess the enhancement potential of sewage sludge for diesel oil bioremediation in soil.

II. MATERIALS AND METHODS

Collection and Processing of Samples

Soil sample used in this study was collected from an agricultural farm land from different sites in Obukpa, Nsukka, Southeast, Nigeria at a depth of 0-30 cm. The soil sample was air-dried for 48 hours and sieved through a 2-mm mesh. Sewage sludge was collected from the University of Nigeria Sewage Treatment Plant while diesel oil was purchased from TOTAL filling station in Nsukka metropolis. Bean seeds (*Phaseolus vulgaris*) were obtained from the Department of Crop Science, University of Nigeria, Nsukka.

Physicochemical Analyses Soil and Sewage Sludge

Physicochemical properties of soil and sewage sludge such as particle size distribution, percentage moisture content, pH, total organic carbon (%TOC), % nitrogen content and total phosphorus were analysed following standard protocol [13].Triplicate determinations were made for each assay.

Determination of Extraction Efficiency of Different Solvents for Diesel Oil

Three different organic solvents namely n-hexane, dichloromethane and diethylether were used to extract diesel oil and their extraction rates were determined. The best solvent in terms of extraction efficiency for diesel oil was later used for the bioremediation assay. The extraction efficiency was determined gravimetrically. Briefly, forty grammes of the soil sample was transferred into a 250 mL flask and polluted with 4 mL of diesel oil. A 4 mL quantity of diesel oil was used so as to simulate a 10% pollution condition that would be studied in the present work. A 100 mL quantity of the three organic solvents was added separately to each polluted soil sample set-up and the set-ups shaken for six hours at 180 rpm. The solution was then filtered using a Whatman No 4 filter paper and the weight of the extracted oil recorded. The extraction efficiency of the organic solvents for diesel was then determined by weight difference following the formula of [14]. The experiment was carried out in triplicates.

$$\text{Extraction efficiency} = \frac{\text{Weight of 4 mL diesel oil} - \text{Weight of oil extracted from soil x 100}}{\text{Weight of 4 mL diesel oil}}$$

Soil Preparation for Bioremediation Study

A 1 kg quantity of the sieved soil was placed in sterile polythene bags and 10 % (v/w) of diesel oil was added, mixed thoroughly, and left undisturbed for 48 hours. After two days, 5%, 10% and 15% (w/w) pulverized sewage sludge were respectively introduced into the diesel oil-polluted soils and mixed thoroughly. Soil sample contaminated with 10% (v/w) diesel oil without sewage sludge amendment served as control. The moisture content of the soil was adjusted to 60% water holding capacity by the addition 50 mL of sterile distilled water (three times weekly) and the set-up kept at room temperature (28±2^{o}C). The experiment was set up in triplicates.

Determination of Diesel Oil Removal from Soil

Periodic sampling from each polythene bag was carried out every seven days in order to determine the residual diesel oil and enumerate heterotrophic microbes. Gravimetric and spectrophotometric methods of [10] with slight modification was employed in the determination of residual diesel oil present in both the control soil and amended options. Composite soil samples weighing five grammes were put in

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

50 mL flasks and 10 mL of n-hexane was added. N-hexane was used because it extracted the highest amount of oil in the extraction efficiency experiment (see result section). The set-ups were shaken with a rotary shaker at 180 rpm for 10 hours to allow for an efficient and complete oil extraction with n-hexane. The mixture was then filtered with a whatman No 4 filter paper. The filtration was done repeatedly two times to ensure complete extraction of the liquid phase. The filtrate was diluted by adding 50 mL of n-hexane to 1 mL of the extracted diesel oil and the absorbance of the solution measured at 460 nm (Shimadzu UV 1800) using n-hexane as blank. The total petroleum hydrocarbon (TPH) was estimated by extrapolating from a standard curve derived from different concentrations of fresh diesel oil diluted with n-hexane. Percent remediation (R) was calculated using the following formula:

$$R = \frac{TPHi - TPHr}{TPH} \times 100$$

Where TPHr and TPHi are residual and initial TPH concentrations

Enumeration and Identification of Oil-utilizing Bacteria and Fungi

Ten-fold serial dilutions of soil samples from each option was made by suspending 10 g of soil in 90 mL of distilled water and shaken vigorously for proper mixing. The suspension was serially diluted up to 10^{-8}. A 0.1mL aliquot of $10^{-5,}$ $10^{-6}$and 10^{-7} dilutions was separately inoculated in sterile nutrient agar plates by the standard spread plate method [15] for active aerobic heterotrophic bacteria isolation. The nutrient agar medium was supplemented with 50 µg/mL nystatin to suppress the growth of fungi. The agar plates were incubated at 37°C for 24 h after which colony forming units per gram of soil samples were calculated. Three replicate samples from each oil-polluted soil were withdrawn every 7 days for the enumeration of total aerobic heterotrophic bacteria (AHB).

Hydrocarbon utilizing bacteria (HUB) in the soil samples were enumerated on Bushnell Has agar using the vapour phase transfer method as described by [16]: A filter paper saturated with sterile diesel oil was aseptically placed on the inside of the cover of inverted inoculated petri dishes and incubated at 28°C for 7 days. Morphologically different colonies of hydrocarbon-utilizing bacteria were picked and pure isolates obtained by repeated sub-culturing on nutrient agar. The bacterial isolates were characterized and tentatively identified using microscopic techniques and biochemical tests such as catalase, urease, oxidase, starch hydrolysis, spore forming, H_2S production, motility, citrate utilization and methyl-red.

For the enumeration and isolation of fungi, 0.1mL of the appropriate dilution of each of the set-ups was inoculated into Sabouraud Dextrose Agar (SDA) plates and incubated at 28±2°C for 4 days. Colony counts were taken and pure isolates obtained by repeated sub-culturing on SDA plates. The fungal isolates were characterized by slide culture and microscopic techniques and identified by the schemes of [17].

Seed Germination Toxicity Test

Toxicity of the soil to seed germination after a 42-day bioremediation experiment was assessed following the seed germination test of [18]. Seeds of *Phaseolus vulgaris* (common bean) were used in this study owing to their sensitivity to hydrocarbon in soil. For each soil preparation, 40 g of thoroughly-mixed remediated soil samples from both the control soils and the amended soil was placed in 100×15 mm petri-dish. Six viable bean seeds were placed evenly throughout each petridish and covered with 10 g of dry sand. The moisture content of the set-ups were maintained at 60% water holding capacity. Triplicate determinations was made for each assay. At the end of 10 days, the number of seeds that germinated from the surface of the soil was counted and root length measured to the nearest centimeter using a metre rule. The results were evaluated using the formula of [19] with slight modification. Soil neither polluted nor amended served as the positive control soil while polluted soil without amendment served as negative control.

- Germination index (%) =(SG×LR)/100
- SG=(ET/CG) × 100
- LR=(LRT/LRC) ×100

  Where SG= number of seed germination, LR=root length (elongation), ET=number of seeds that germinated on treated soil, CG=number of seeds that germinated on positive control soil, LRT= root length on treated soil, LRC=root length on positive control soil.

Statistical Analysis of Data

The data obtained in the present study were subjected to one-way analysis of variance (ANOVA). Relationship between variables and comparison of means of the different treatments were tested for level of significances at $P \leq 0.05$ using least square difference and post-hoc multiple comparison tests. The data analysis was performed using SPSS.

III. RESULTS

Physicochemical Properties of Soil and Sewage Sludge

The physicochemical properties of the soil and SS used in this study are presented in **Table 1**. The % organic nitrogen

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

content of the soil was 0.02 and available phosphorus content of 10.64%. Other parameters of the soil in percentage are: TOC 2.49, moisture 15.38 and the pH is 4.9. The sewage sludge had pH, nitrogen content, TOC and total phosphorus content of 3.4±0.015, 0.28±0.03, 11.97±0.07 and 17.92±0.034, respectively and nitrogen content of 0.28 and TOC of 11.97. Pig droppings had nitrogen content of 0.238 while poultry manure had organic nitrogen content of 0.098 and TOC of 56.86

Table 1. Physicochemical Properties of Soil and Sewage Sludge

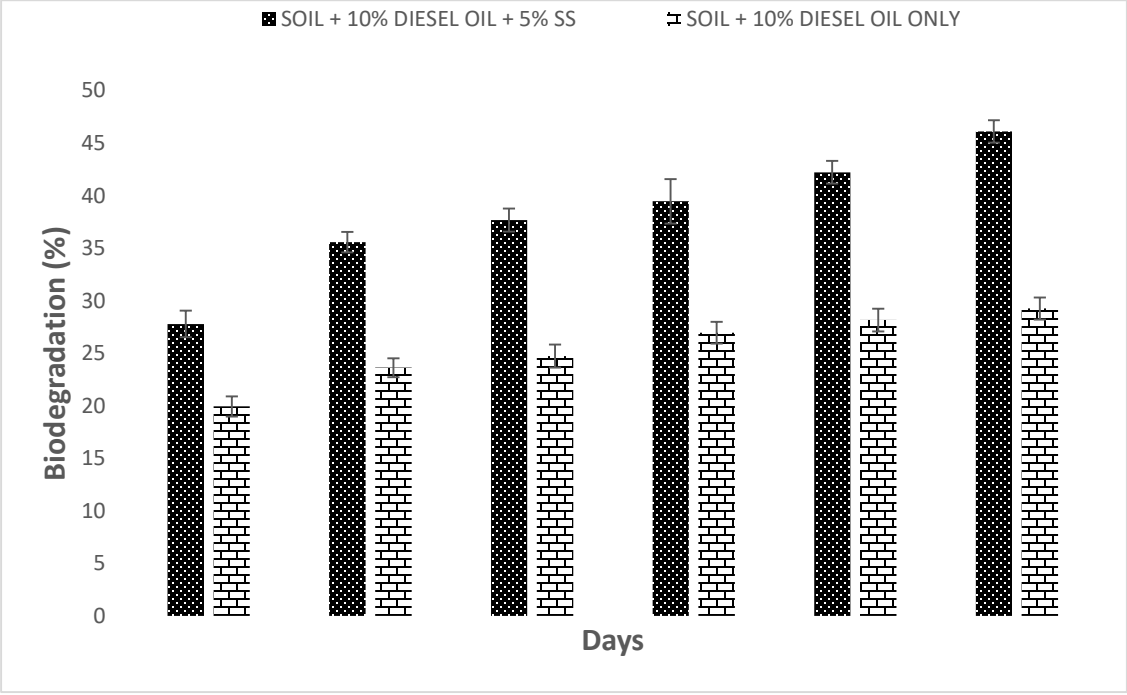
Parameter	Non-polluted Soil	Sewage sludge
pH	4.90±0.37	3.40±0.015
Nitrogen	0.042±0.02	0.28±0.030
Organic carbon	2.49±0.45	11.97±0.07
Phosphorus (PPM)	10.64±0.50	17.92±0.034
Moisture(%)	10.38±0.30	8.69±0.229
Clay (%)	71.00 ± 0.41	-
Silt (%)	19.50± 0.06	-
Sand (%)	9.50± 0.08	-
Texture	Clayey Loam	

Extraction Efficiency of Solvents for Diesel Oil

The amount of diesel oil (in percentage) that was extracted by three different solvents namely: n-hexane, dichloromethane and diethylether six hours after polluting soil with 10% (v/w) diesel oil were 80.72%, 80.68%, 80.56%.

Bioremediation of diesel oil in soil

The levels of bioremediation of diesel oil in the unamended control soil and soil amended with 5% SSare shown in Figure1. Percentage oil loss in the sewage sludge-amended option ranged between 27.78±1.27 and 58.33±0.67. In the unamended control soil, the percentage loss ranged between 19.93±0.96 and 32.22±0.17



https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

Fig.1: Bioremediation of diesel oil in polluted soil amended with 5% SS

Figure 2 shows the level of oil loss in control soil and polluted soil amended with 10% SS over a 42-day period. Percentage oil loss in the SS-amended option ranged from 35.94±0.98 to 48.75±1.27

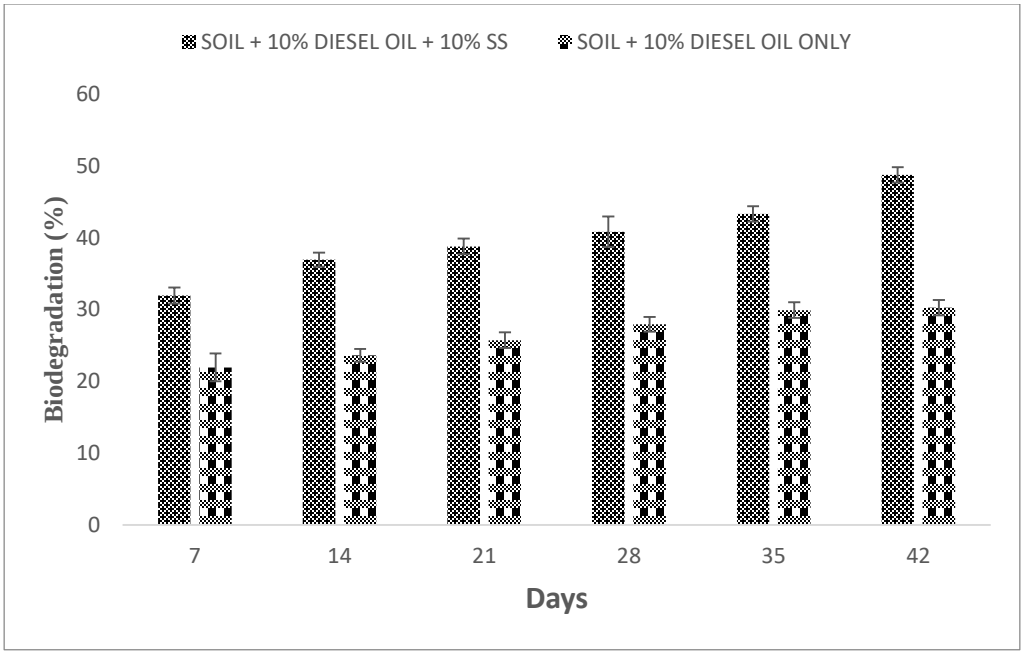
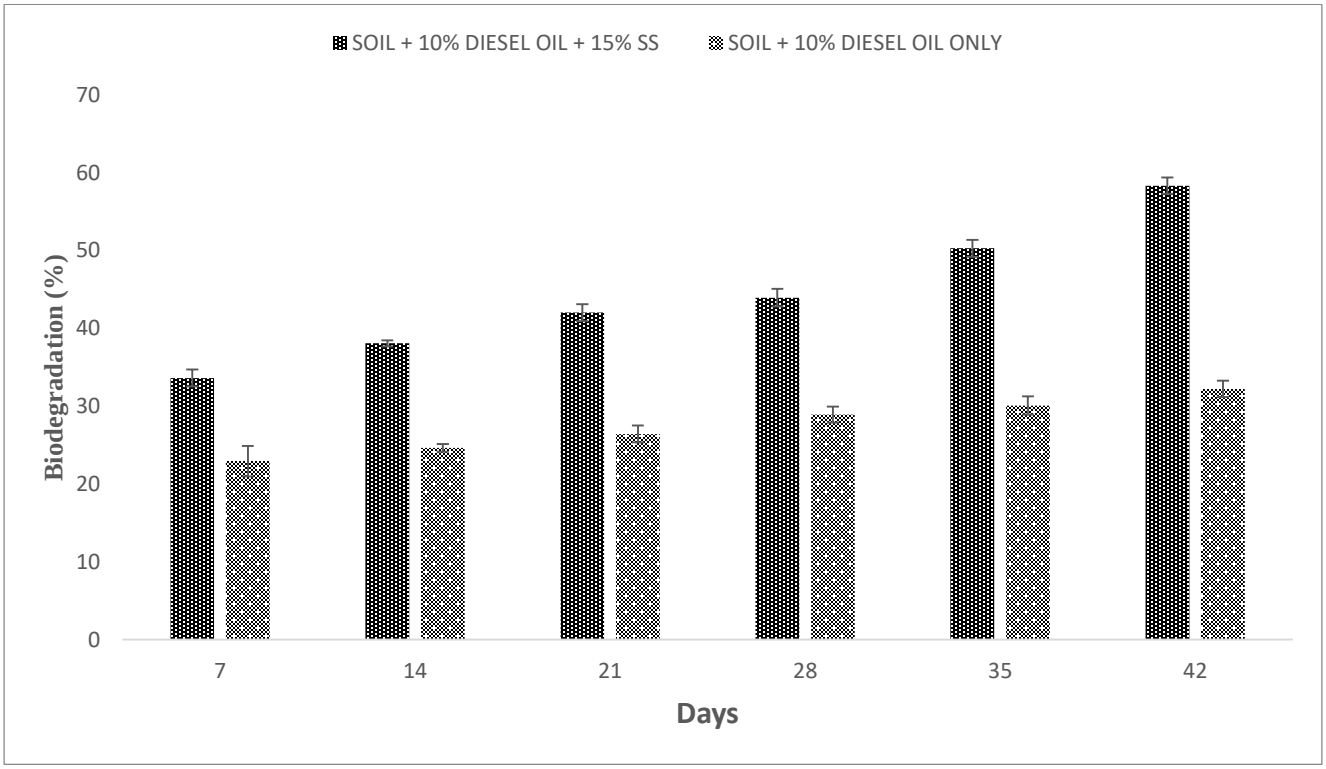


Fig.2: Bioremediation of diesel oil in polluted soil amended with 10% SS

The level of oil loss in the control soil and polluted soil amended with 15% SS over a 42-day period is presented in **Figure 3**. Percentage oil loss in the SS-amended option ranged from 36.61±1.09 to 58.33±2.12



https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

Fig.3: Bioremediation of diesel oil in polluted soil amended with 15% SS

Active Aerobic Heterotrophic Bacterial (AHB) Counts

The total heterotrophic bacteria in control soil and polluted soil amended with 5% SS are presented in **Figure 4**. AHB counts in the SS-amended option ranged from 8.3 x 10^7±2.78 to 22.5 x 10^7 ±0.71 CFU/g. Control soil had AHB counts ranging from 1.0 x 10^7±0.6 to 20 x 10^7±0.9

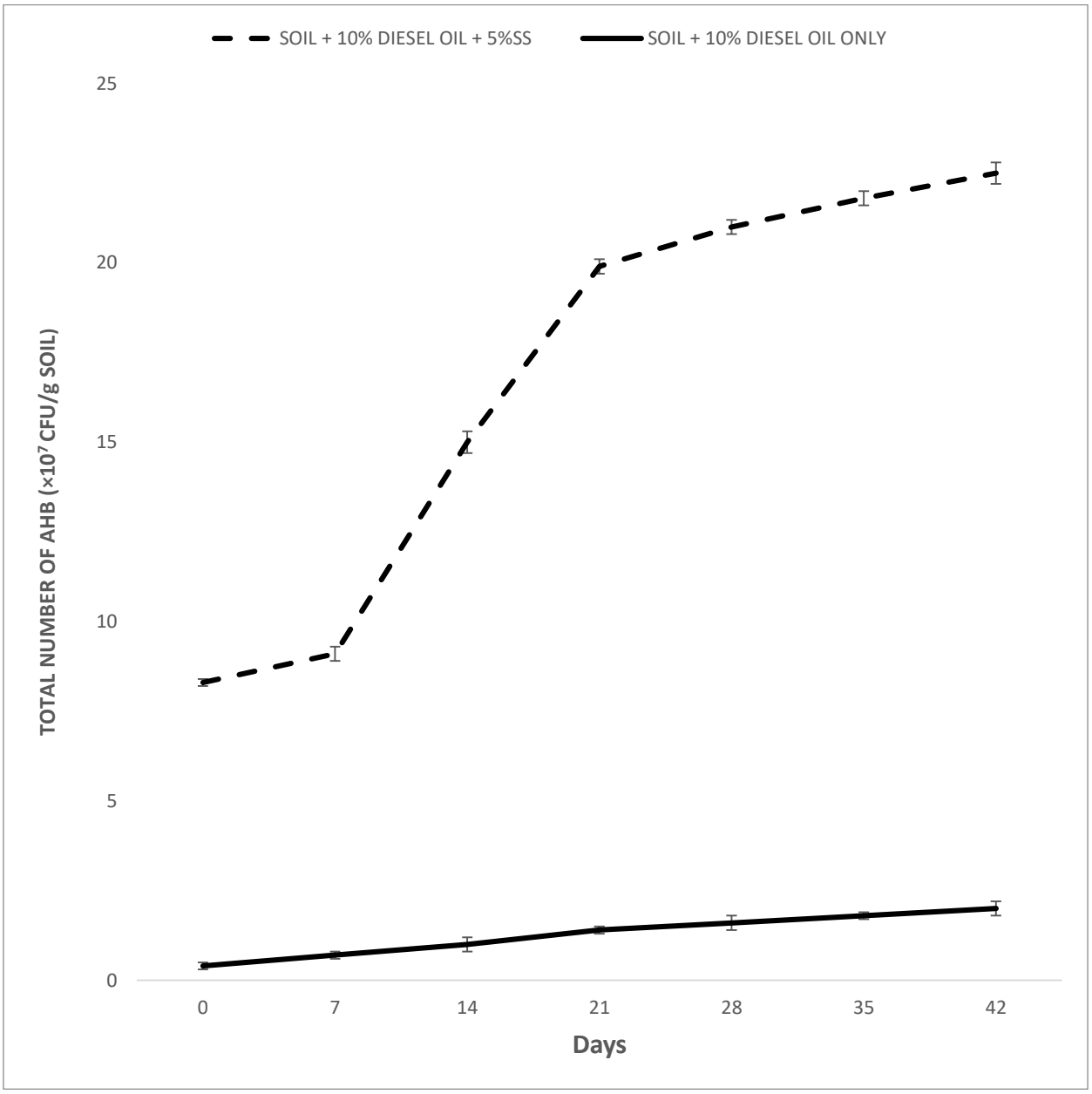


Fig.4: Aerobic heterotrophic bacterial (AHB) population in soil contaminated with 10% diesel oil and amended with 5% SS

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

Figure 5 shows the total number of aerobic heterotrophic bacteria in polluted soil amended with 10% SS and the control option. AHB in SS-amended option ranged from 9.2 x 10^7 ±0.28 to 23.3 x 10^7 ±0.14 CFU/g

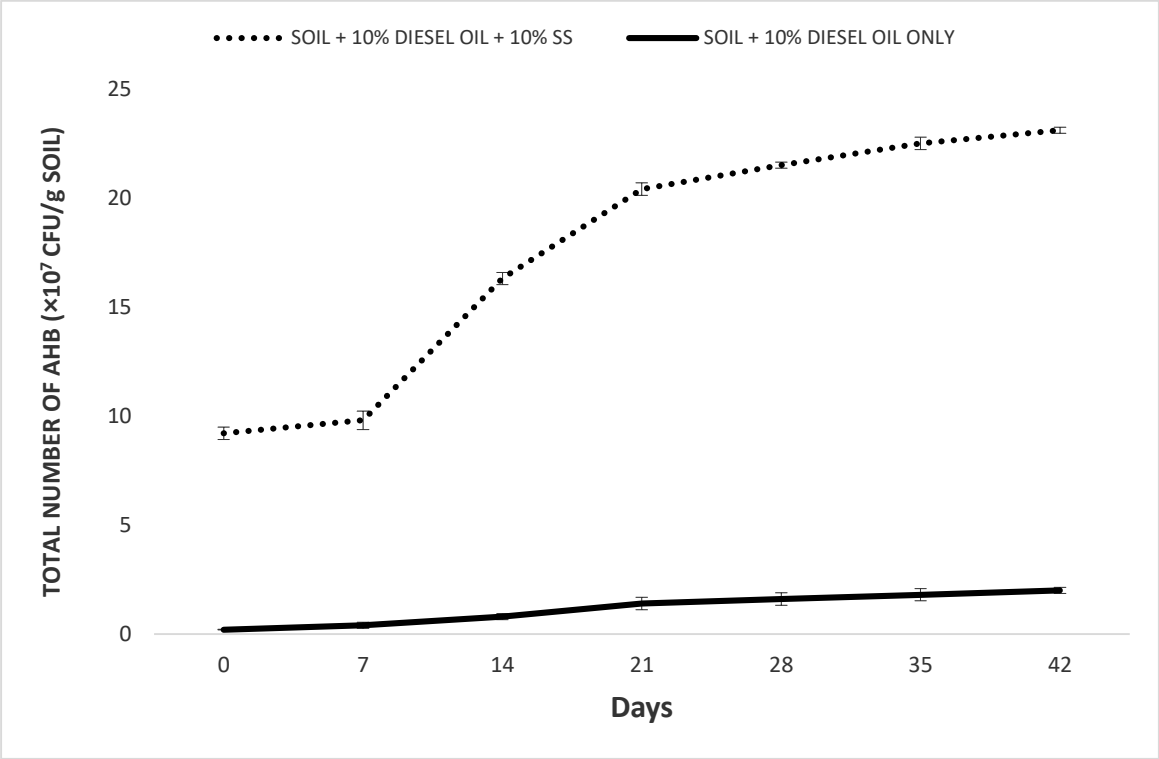


Fig.5: Aerobic heterotrophic bacterial (AHB) population in soil contaminated with 10% diesel oil and amended with 10% SS

AHB counts in the control option and polluted soil amended with 15% SS are presented in **Figure 6.** AHB counts ranged from 10.5 x 10^7 ±0.28 to 24.0 x 10^7 ±0.78 in the SS-amended option.

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

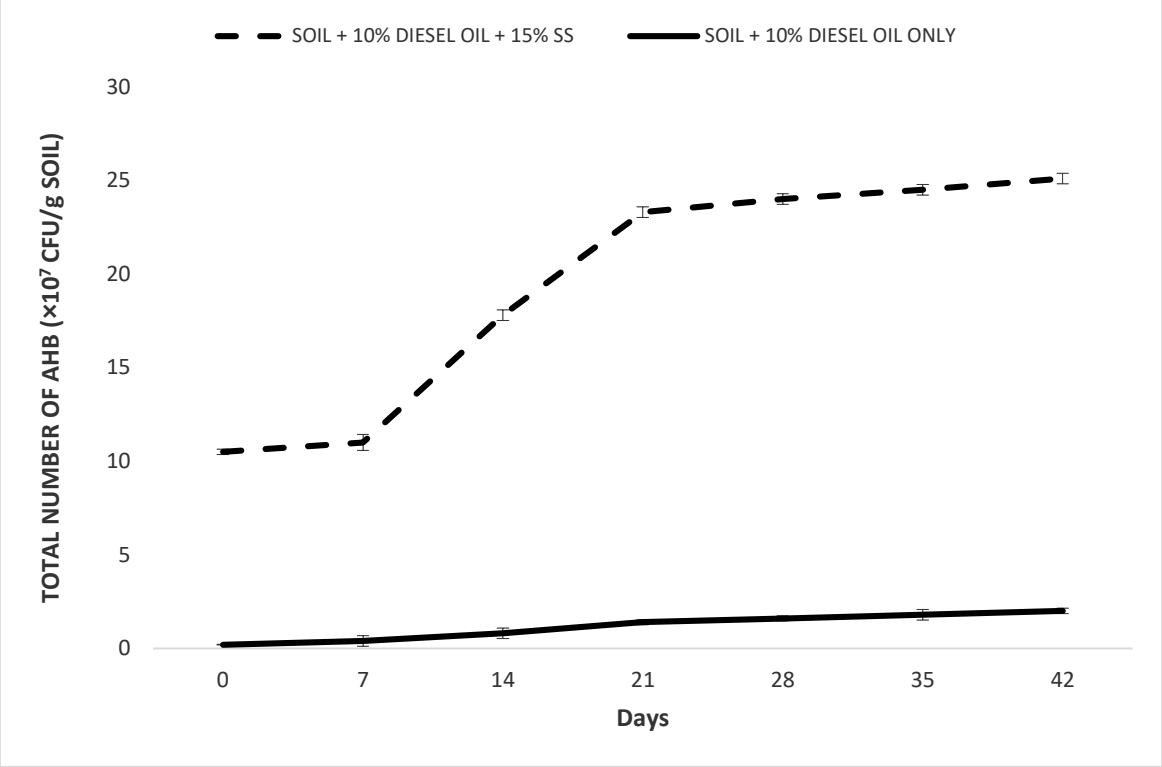


Fig.6: Aerobic heterotrophic bacterial (AHB) population in soil contaminated with 10% diesel oil and amended with 15% SS

Hydrocarbon-Utilising Bacterial (HUB) Counts

The profile of HUB count in control soil and oil-polluted soil amended with 5% SS over a 42-day period is presented in **Figure 7**. HUB counts in the SS-amended option ranged from 4.8 x 10^6±0.71 to 11.0 x 10^6 ±0.78. The control option had a HUB count ranging from 0.8 x 10^6±0.13 to 3.2 x 10^6± 0.27.

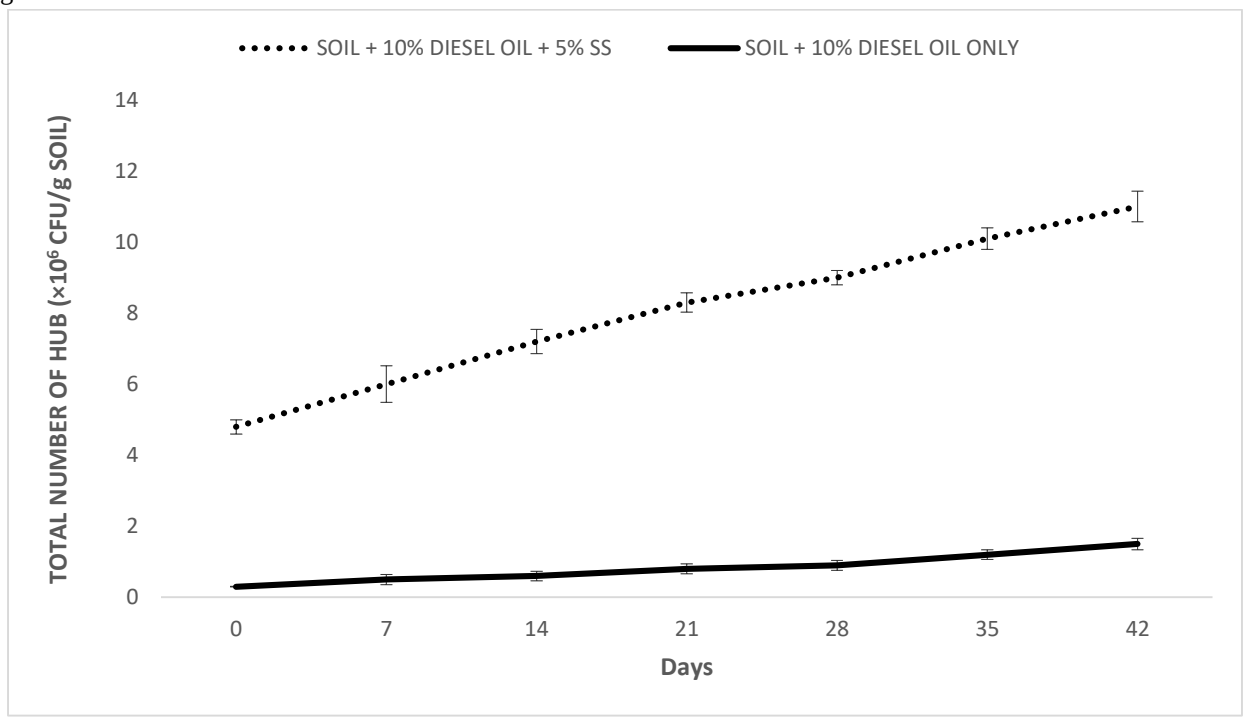


Fig.7: HUB counts in soil contaminated with 10% diesel oil and amended with 5% SS

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

Figure 8 presents the HUB counts in the 10% SS-amendment option over a forty-two day period. HUB counts in the SS-amended option ranged from 5.0 x 10^6 ±0.48 to 12.0 x 10^6 ±0.81

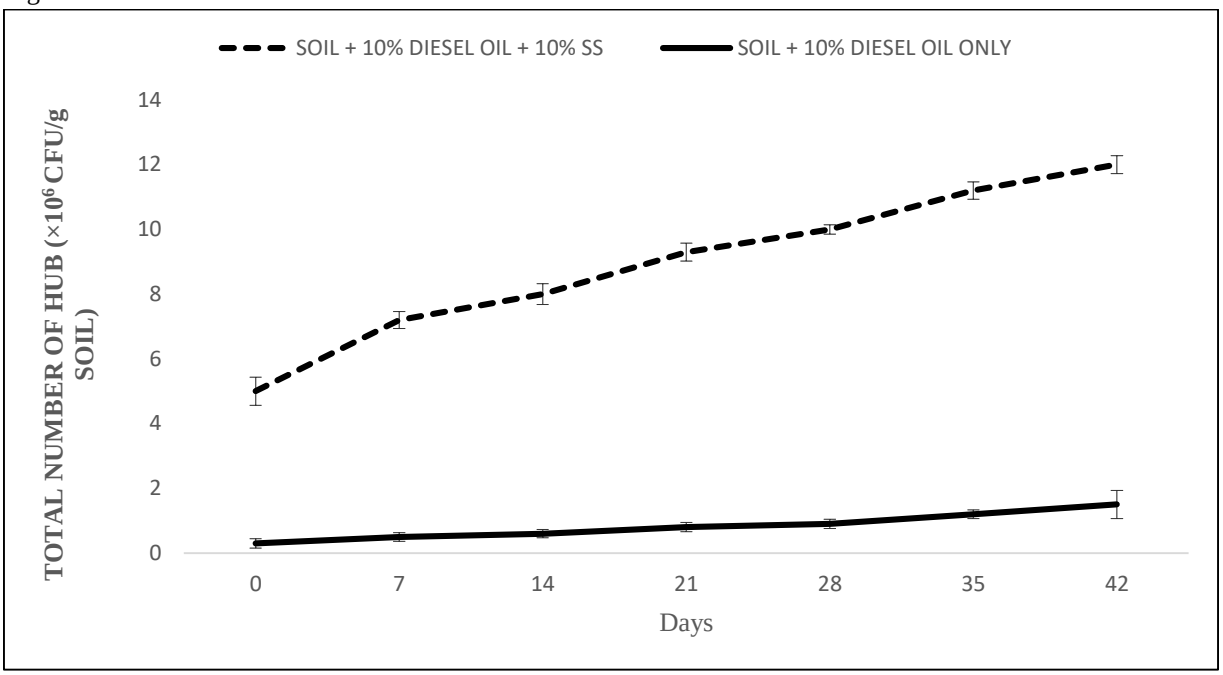


Fig.8: HUB counts in soil contaminated with 10% diesel oil and amended with 10% SS.

The hydrocarbon-utilising bacterial load in the control option and oil-polluted soil amended with 15% SS within 42 days are presented in **Figure 9.** AHB ranged from 6.2 x 10^6 ±0.31 to 12.2 x 10^6 ±0.48 in the SS-amended option.

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

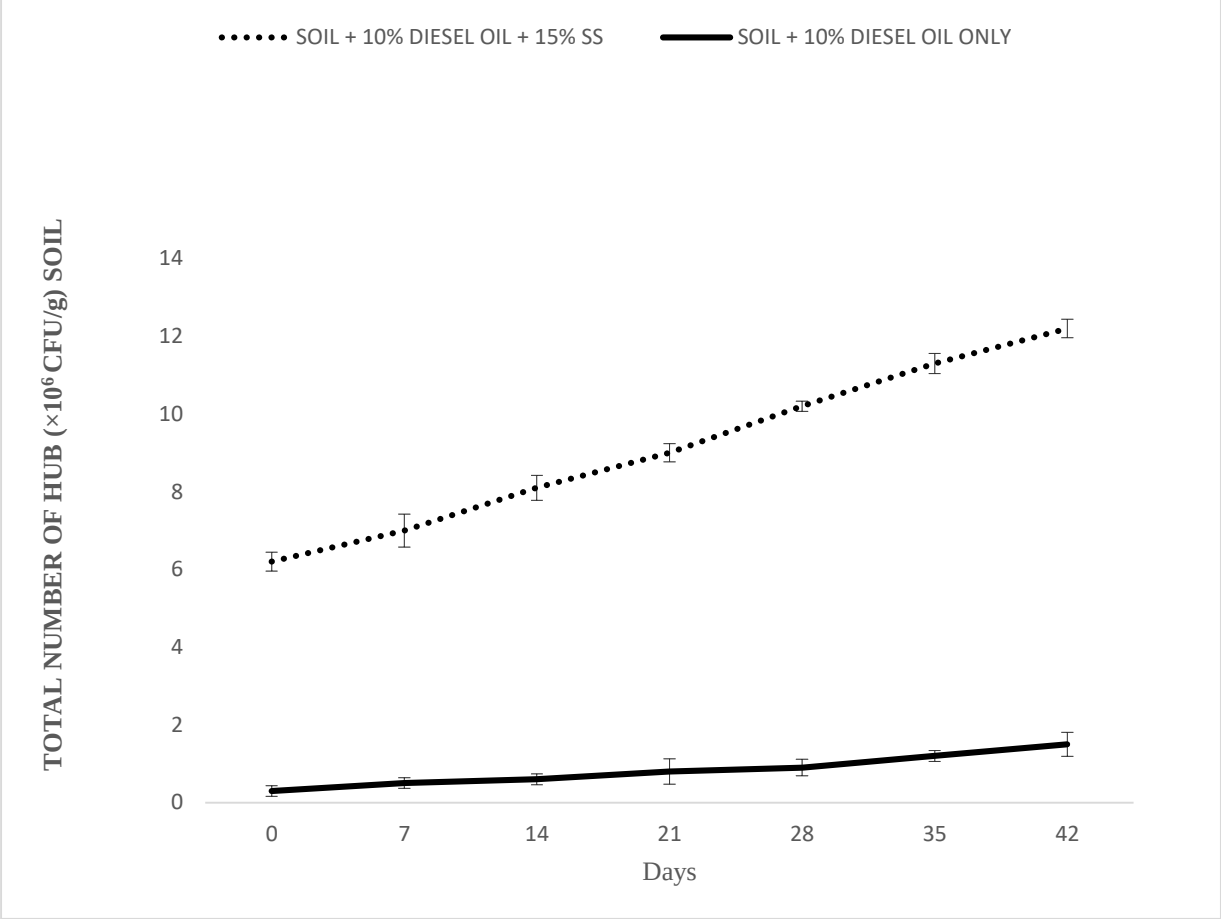


Fig.9: HUB counts in soil contaminated with 10% diesel oil and amended with 15% SS.

Identities of Bacterial isolates

The microscopic and biochemical characteristics of the isolated hydrocarbon-utilising bacteria are presented in **Table 2**. The HUBs are identified tentatively as *Bacillus licheniformis*, *Pseudomonas putida*, *Corynebacterium* sp., *Micrococcus varians*, *Acinetobacter sp and Bacillus cereus*

Table 2:Microscopic and Biochemical Characteristics of Bacterial Isolates

Gram reaction	Catalase test	Oxidase test	H_2S	Starch hydrolysis	Methyl red	Urease test	Citrate utilisation	motility	Spore-formation	Probable identity
+	+	-	-	+	+	-	-	+	+	*Bacillus cereus*
+	+	+	+	-	+	+	+	-	-	*Micrococcus varians*
-	+	-	+	-	-	-	+	-	-	*Acinetobacter* sp
-	+	+	-	+	-	+	+	+	-	*Pseudomonas putida*
+	+	+	-	-	-	-	-	-	-	*Corynebacterium* sp
+	-	-	-	+	-	-	-	+	+	*Bacillus licheniformis*

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

Fungal Counts

The active aerobic heterotrophic fungal counts in the 5% amendment options and unamended control soil over a 42-day period is presented in **Figure 10.** Fungal counts in the SS-amended soil ranged from 3.8 x 10^5 ±0.11 to 9.6 x 10^5 ±0.38 while 0.3-2.0 x 10^5 ±0.13-0.23 is the range of fungal counts recorded in the unamended control soil.

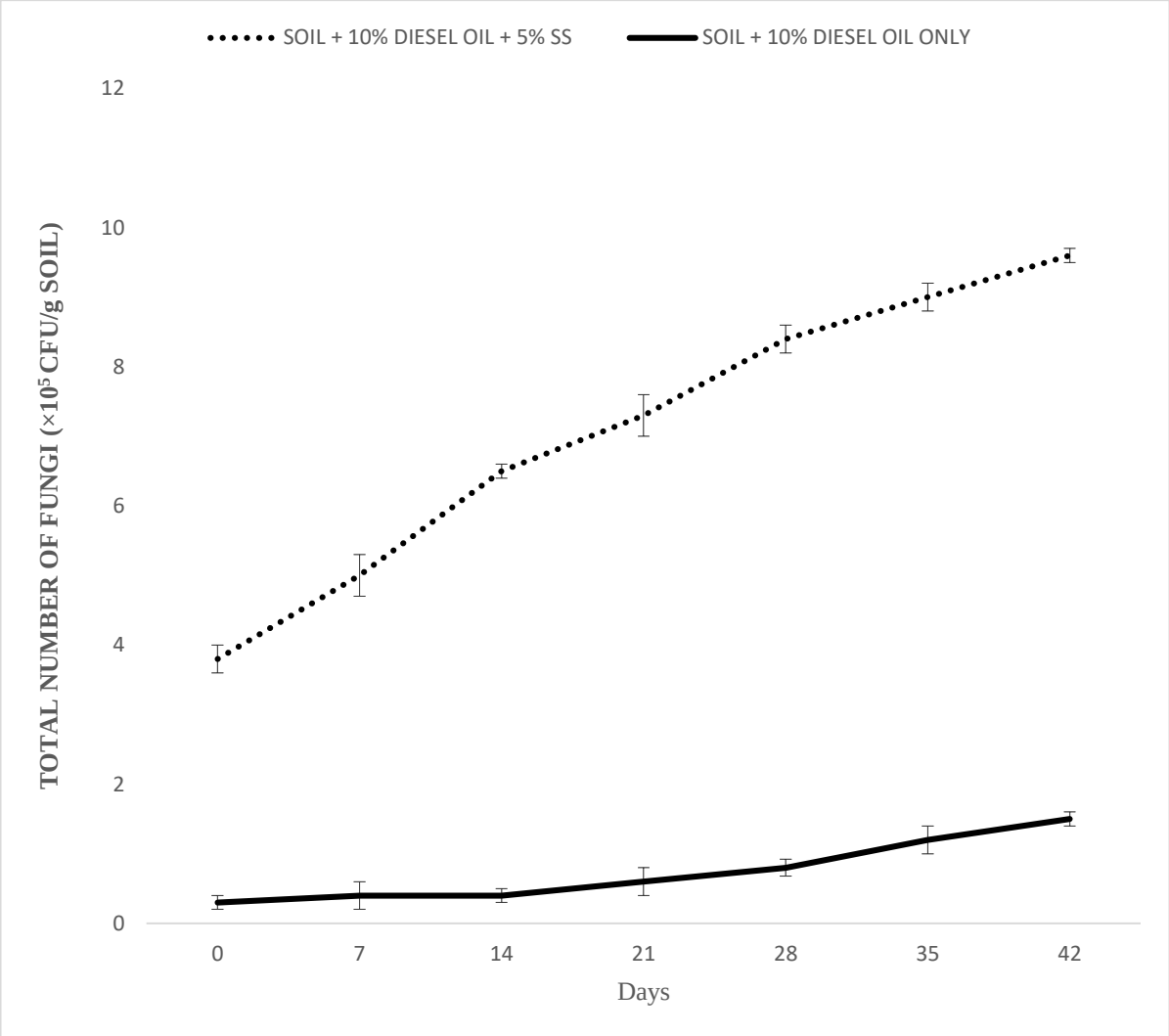


Fig.10: Fungal population in soil contaminated with 10% diesel oil and amended with 5% SS.

Fungal counts in the oil-polluted soil amended with 10% SS and the control option over a 42-day period is presented in **Figure 11.** Fungal counts ranged from 4.9 x 10^5 to 10.9 x 10^5 in the SS-amended soil.

https://dx.doi.org/10.22161/ijeab.51.23 ISSN: 2456-1878

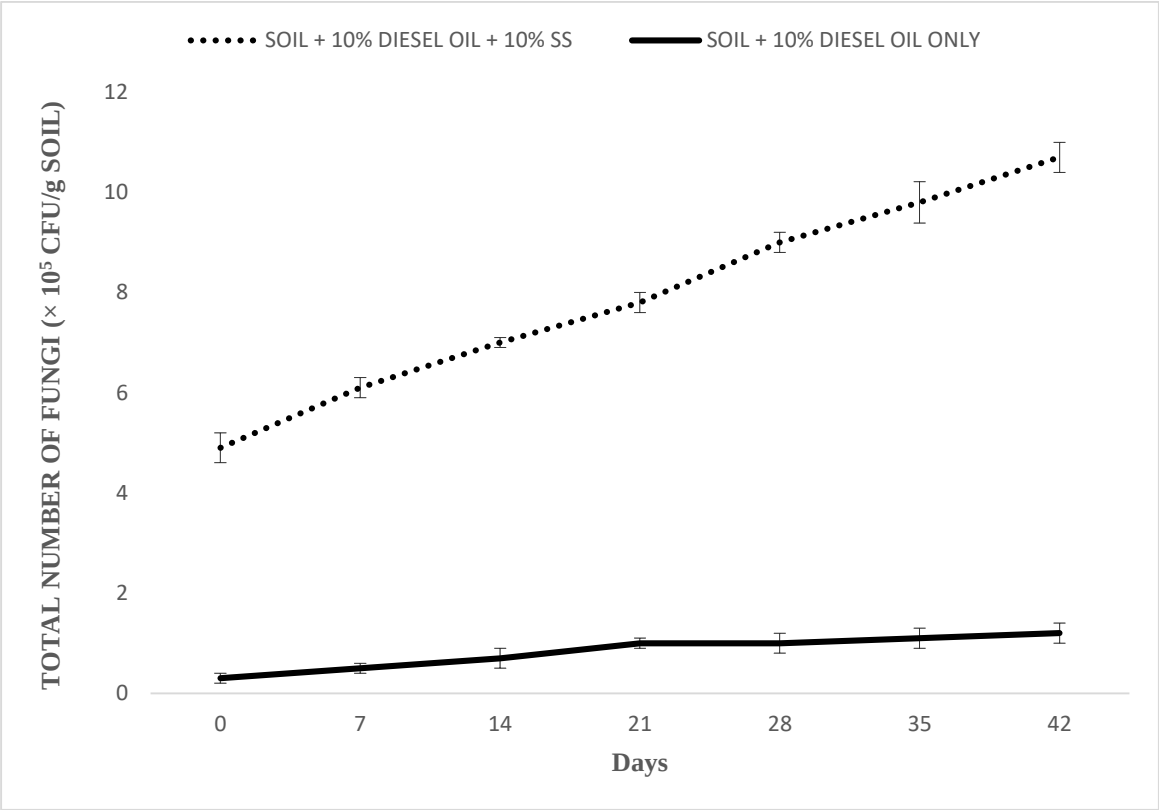


Fig.11: Fungal population in soil polluted with 10% diesel oil and amended with 10% SS.

Fungal load recorded in the 15% SS-amendment options over a 42-day period is presented in **Figure 12.** It ranged from 6.0 x 10^5 ±0.22to 11.6 x 10^5 ±0.39

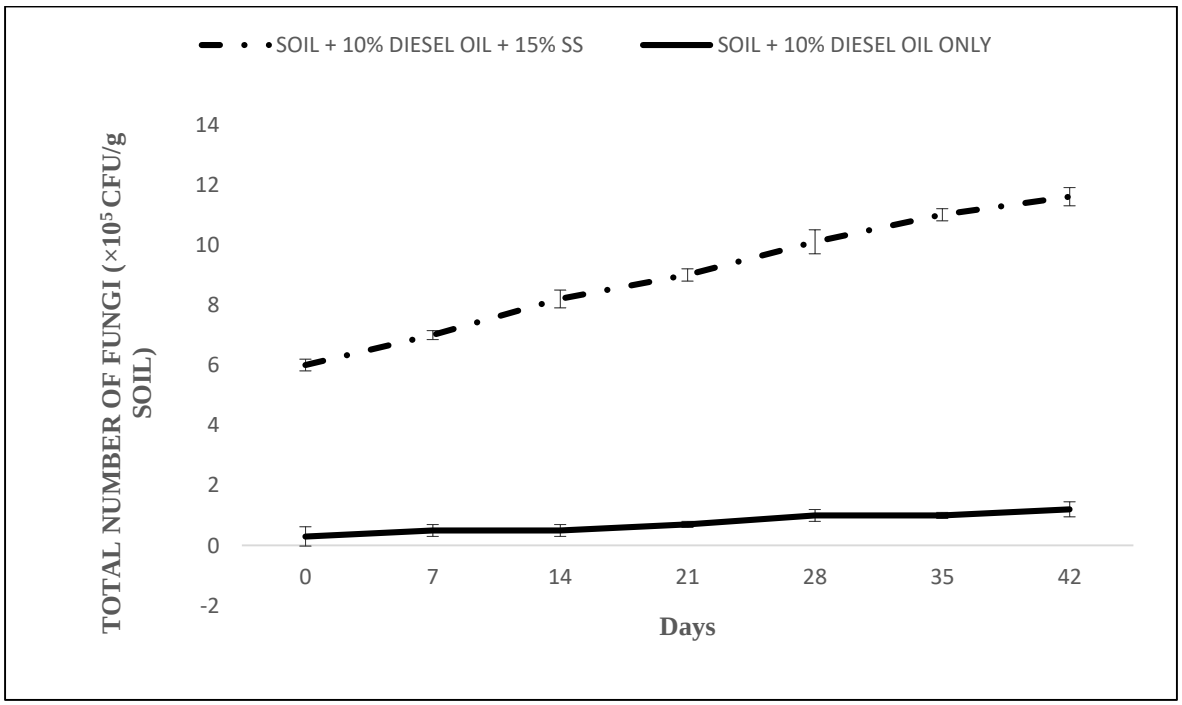


Fig.12: Fungal population in soil polluted with 10% diesel oil and amended with 15% organic wastes.

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

Identities of Fungal Isolates

The cultural and microscopic characteristics of the isolated hydrocarbon-utilising fungi are presented in Table 3. Fungi isolated predominantly were identified tentatively as *Aspergillus niger, Aspergillus flavus, Fusarium* sp., *Cladosporium* sp. *and Penicillium* sp

Table 3: Cultural and Microscopic Characteristics of Fungal Isolates

Cultural characteristics	Microscopic Characteristics	Probable Identity
Dark brown, powdery, flat spread on the surface of the solid medium with reverse	septate and branched hyphae with conidia in chains	*Aspergillus* niger
Yellow, powdery, flat spread on the surface of the solid medium with colourless reverse	Septate and branched hyphae with conidia in chains	*Aspergillus* flavus
Grey colonies that were large with white border. Colourless or white reverse	Long conidiophores consisting of broom-like conidia in chains	*Penicillium* sp
Whitish and cottony mycelium with pinkish pigments at the centre. Brown reverse side	segmented canoe-shaped spores and branched conidiophores	*Fusarium* spp
Powdery, slow-growing, blackish-brown colonies, olivaceous-black reverse. Conidiophores and conidia equally pigmented	Branched and shield-shaped conidia in chains	*Cladosporium* sp

Seed Germination Toxicity

Seed germination parametres employed in this study are presented in **Table 4**. SG (%) ranged from 66.7 to 83.3 across all amendment levels in the SS-amendment option while negative and positive control soils had SG (%) of 16.7 and 100, respectively. LR (%) ranged from 58.5 to 87.7 across all amendment levels in the amendment option while 16.9 and 100 were recorded in the negative and positive control, respectively. Furthermore, GI ranged from 39.0 to 73.1 across all amendment options. Negative and positive control had GI of 2.8 and 100

Table 4: Seed Germination Parametres

Soil preparations	SG	LR(cm)	SG(%)	LR(%)	GI
5% SS	4.0±0.14	3.8±0.43	66.7	58.5	39.0
10% SS	5.0±0.43	4.5±0.15	83.3	69.2	57.6
15% SS	5.0±0.26	5.7±0.02	83.3	87.7	73.1
Negative control	1.0±0.00	1.1±0.01	16.7	16.9	2.8
Positive control	6.0±0.01	6.5±0.00	100	100	100

Key: number of seeds that germinated, LR= root length, GI= germination index

IV. DISCUSSION

Several studies on hydrocarbon remediation have shown that bioremediation employing the activities of microbes alone proved inadequate [20, 11, 12]. It was argued [21] that while hydrocarbons are excellent sources of carbon and energy to microbes, they are incomplete foods in that they do not contain significant concentrations of other nutrients such as nitrogen and phosphorus required for microbial growth. The dearth of nutrients in soils polluted with hydrocarbons poses a challenge to bioremediation; nevertheless, nutrient addition generally favours soil hydrocarbon-utilising bacteria and fungi, ultimately resulting in enhanced bioremediation of hydrocarbon-polluted environments [8]. By adding organic wastes, the C:N and C:P ratios of the soil becomes closer to the bacterial requirements of the same.

It was reported [3] that bioremediation of diesel fuel depended on phosphorus ability. However, some nutrient sources might supply enough phosphorus to restore the microbial C:P relationship but become unavailable due to their low solubility [22]. Pivotal therefore, is the knowledge of nutrient bioavailability to planning an efficient bioremediation protocol.

The percentage nitrogen content of the soil used in this study was comparably low **(Table 1)**. Hydrocarbon degraders need more nitrogen and phosphorus than is normally present in soils to convert excess carbon present in hydrocarbon

pollutants to biomass [21]. In a similar vein, it was stated [23] that although microorganisms are present in contaminated soil, they cannot necessarily be present at levels required for bioremediation of the site, hence the need for their growth and activities to be stimulated. The soil used in this study had high C:N ratio of 59:1 **(Table 1)**. Furthermore, pollution of the experimental soil with 10% v/w diesel oil introduced more carbon to the soil, thereby increasing its C:N ratio. As a general rule of thumb, materials with a C:N ratio greater than 25:1 stimulate immobilization [24]. Carbon is the most basic form of nutrient required for living organism. In addition to this, bacteria also need macronutrients such as nitrogen and phosphorus to ensure effective degradation of the oil. However, the ratio of carbon to essential nutrients such as nitrogen and phosphorus is critical to the realization of an effective bioremediation. As organisms convert excess carbon present in hydrocarbon spills onto biomass, they require corresponding proportions of these essential nutrients. The optimum nutrient balance required for hydrocarbon remediation is C:N:P equals 100:10:1 [25,23]. Higher polycyclic aromatic hydrocarbon degradation was recorded in soil amendment with C:N ratio of 10:1 than those with C:N ratio of 25:1 and 40:1, respectively [26]. In the present study, therefore, the addition of sewage sludge rich in organic nitrogen obviously balanced the C:N ratio of the soil at a level that enhanced growth and activities of microbes. This enhancement ultimately favoured degradation of diesel oil pollutant.

The result of the extraction efficiency experiment clearly indicated that n-hexane was the best choice in extracting diesel oil under the conditions employed in this study. This is due to the fact that the highest amount of diesel oil was extracted with n-hexane among other solvents such as dichloromethane and diethylether used in this study.

Percentage oil loss (bioremediation) increased appreciably from the first week to the sixth week in both the amendment and control options (**Figures 1-3**). However, as was observed throughout the study period, oil loss increased with increasing concentration of sewage sludge. Highest oil loss was noted in the polluted soil amended with 15% SS (**Figure 3**). The observation of highest oil loss in the SS-amended option was probably due to its relatively higher content of organic nitrogen (**Table 1**). Positive effects of nitrogen amendment has been demonstrated [27]. It has been reported that when oil is applied at rates of 0.5-10% based on the weight, extensive bioremediation of the oil components occurs within the first three months [12]. Oil loss in the control option also increased notably from 19.23% to 32.22% at the end of 42 days. A similar study on diesel oil remediation using soy cake, potato skin and tea leaf amendment recorded 35% oil loss in the control option [28]. A 42-day study on crude oil remediation using only goat manure as amendment recorded 8.15% oil loss for the control microcosm at the end of the study [16]. Part of the oil loss in the control option could be due to some factors such as natural bio attenuation by the indigenous hydrocarbon-degrading flora, photo degradation, volatilization, sorption, dilution and dispersion. Similar observation was noted [22]. Greater oil loss was recorded in the amendment options than the control option. Similar trend have been widely documented [10, 12, 22].

Even though the application of bio stimulation strategy in bioremediation has gained wide acceptance, reports on the bio stimulation potentials of organic amendments have been widely divergent in literature. While some researchers documented a direct (linear) relationship between hydrocarbon remediation and impact of organic wastes [8,29], another proved otherwise by reporting that natural attenuation was more successful than bio stimulation in Hong Kong soil [30]. It was also found that nutrient supplementation had no significant effect on the remediation of polluted soils [31]. However, it was asserted that different soils have varying inherent microbial potentials to degrade hydrocarbons [32]. Ways to activate these potentials must bring into account that most degradation potentials are widely distributed among microorganisms and indigenous microbes are always present in small numbers.

The growth and activities of heterotrophic bacteria and fungi are a biological indicator of the impact of organic wastes. In the present study, AHB counts increased progressively throughout the study in the amendment and control options **(Figures 4-6).** However, AHB counts were higher in the amended option (at all amendment levels) than the control option. It was also observed that highest counts were recorded at 15% amendment level. This could be as a result of enhanced nutrient level in the highest level of amendment. Earlier researchers noted similar observation [11, 33]. AHB counts of 8.0×10^6-30.0×10^6 was recorded in the amended option in a study on diesel oil remediation using cowpea chaff [33]. In a similar study, AHB counts ranging from 3.4×10^3-2.9×10^5CFU/g were recorded in the control soil [16]. Similarly, HUB counts increased markedly from $4.8 \times 10^6 \pm 0.9$ to $12.3 \times 10^6 \pm 0.75$ CFU/g soil throughout the study period **(Figures 7-9)** and higher HUB counts were recorded in the amended option than the control. Similar observation had been documented [34]. Furthermore, as was noted in the

https://dx.doi.org/10.22161/ijeab.51.23 *ISSN: 2456-1878*

population count profile of AHB and HUB, fungal population increased progressively throughout the study period (**Figures 10-12)** and was highest at the highest amendment level (15%) (**Figure 12).** The observation of greater AHB, HUB and fungi in the amended option than the control option might also be attributed to the fact that sewage sludge harbor great diversities of microbes with inherent hydrocarbon-degrading potentials. Technically speaking therefore, addition of organic wastes such as sewage sludge can be regarded as 'uncontrolled bioaugmentation'. It was noted in this study that AHB counts were higher in number than their hydrocarbon-utilising counterparts. Similar trend was noted [20]. It stands to reason therefore that HUB are a significant proportion of heterotrophic bacteria that evolved probably as a result of incessant soil pollution with hydrocarbons. The HUB isolated in this study were identified tentatively as *Bacillus licheniformis*, *Pseudomonas putida*, *Corynebacterium* sp., *Micrococcus varians*, *Acinetobacter* sp *and Bacillus cereus* (**Table 2)**. These bacteria have been widely reported [11, 20, 22] as having hydrocarbon-utilisation attributes. Also, fungi isolated in the present study were identified tentatively as *Aspergillus niger, Aspergillus flavus, Fusarium* sp., *Cladosporium* sp. *and Penicillium* sp. (**Table 3)** These have also been reported by several researchers [29, 35] as being implicated in hydrocarbon degradation. It was observed that the hydrocarbon-degrading bacterial population in the amended option were higher than their fungal counterparts. It was argued [36] that although it is widely accepted that bacteria and fungi are primary mediators in hydrocarbon remediation, bacteria have been found to be more versatile than fungi and therefore may play a greater role during biodegradation of hydrocarbons. Generally, bioremediation levels and microbial counts may vary among similar studies. This may be due to the assertion that the fate and effect of oil depends on the type of oil and extent of pollution, properties of oil as modified overtime by physical and chemical processes, the organisms and habitats exposed and the nature of the exposure [21].

Seed germination toxicity test has been demonstrated as a good parameter of assessing the efficacy of bioremediation on contaminated soils [18]. The highest germination index was recorded in the SS-amended option at 15% amendment level (**Table 4**). GI recorded in the present study followed the same pattern as bioremediation results and microbial counts. All the seeds germinated in the positive control soil while one seed germinated in the negative control soil (**Table 4**). This could be attributed to absence of oil pollution in the positive control soil and oil pollution in the negative control soil. Oil pollution has been identified by different researchers [37, 2] as having adverse effects on plant development parametres. Growth of all seeds of *Moringa olifera* planted in the positive control option was recorded [38]. Positive control option also allowed 99.6% germination [37]. *Phaseolus vulgaris* normally germinates within 8-10 days but the germination was delayed to 19-21 days owing to slightly heavy pollution simulated in this study (10%) which had not been fully remediated (58.33%) as at the end of the 42-day study period. There was a significant difference in bioremediation level and microbial counts ($P \leq 0.05$) between SS-amended soil and control even at 5% level.

V. CONCLUSION

This study showed that bioremediation employing the impact of organic amendments to soil polluted with diesel oil enhanced the activities of the remediating microflora by improving nutrient supply. Sewage sludge alone or in combination with other wastes can therefore offer a good alternative to the rather expensive physical and chemical methods of hydrocarbon remediation.

ACKNOWLEDGEMENT

The authors' appreciation goes to Dr. V.N. Chigor of the Department of Microbiology, University of Nigeria for the use of his laboratory facilities.

REFERENCES

[1] APHA (2008) American Public Health Association, Standard Methods for the Examination of Water and Wastewater, 21st Edition, Washington DC.

[2] Ayandele, A.A. (2018). Biotreatment of soil contaminated with spent engine oil by locally isolated microorganisms. *Advances in Environmental Microbiology*. 12(11):22-28

[3] Ogbo, E.M. (2009). Effects of diesel fuel contamination on seed germination of four crop plants-*Arachis hypogaea, Vigna unguiculata,Sorghum bicolor,* and *Zea mays*. African Journal of Biotechnology. 8:(2):250-253

[4] Agarry, S.E. and Latinwo, G.K. (2015). Biodegradation of diesel oil in soil and its enhancement by application of bioventing and amendment with brewery waste effluents as biostimulation bioaugmentation agents. *Journal of Ecological Engineering*.16(2):82-91

[5] Brady, R.N.(2001). Diesel Fuel. In:*Macmillan Encyclopedia of Energy*. Vol. 1. Ed. John Zumerchik. Macmillan Reference USA. New York: 336-342

[6] Riffaldi, R., Levi-Minzi, R., Cardelli, R., Palumbo, S., Saviozzi, A.(2006). Soil biological activities in monitoring the bioremediation of diesel oil-contaminated soil. *Water, Air,and Soil Pollution*,170, 3-15.

[7] Eramo, A. (2009).Bioremediation of diesel contaminated soil using spent mushroom compost. A thesis in Environmental Engineering.The Pennsylvania State University. Pp 1-3

[8] Awofeso, N.(2011). Generator diesel exhaust: a major hazard to health and the environment in Nigeria. *American Journal of Respiratory and Critical Care Medicine*, 183(10), pp. 1437.

[9] Abioye, P., Abdul Aziz, A., Agamuthu, P. (2009a). Enhanced biodegradation of used engine oil in soil amended with organic wastes. *Water, Air, and Soil Pollution*, 209(1): 173-179.

[10] Azubuike, C.C., Chikere, C.B. and Okpokwasili, G.C. *World Journal of Microbiology and Biotechnology* (2016) 32: 180. https://doi.org/10.1007/s11274-016-2137-x. Retrieved 05/12/19

[11] Adesodun J.K., Mbagwu J.S.C. (2008), Biodegradation of waste-lubricating petroleum oil in a tropical alfisol as mediated by animal droppings, *Bioresource Technology*, 99(13), 5659-5665.

[12] Onuoha, S.C. (2013).stimulated biodegradation of spent lubricating motor oil in soil amended with animal droppings**.** *Journal of Natural Sciences Research*,3 (12), 106-116

[13] APHA (2008) American Public Health Association, Standard Methods for the Examination of Water and Wastewater, 21st Edition, Washington DC.

[14] Agamuthu, P., Dadrasnia, A. (2013). Potential of biowastes to remediate diesel fuel contaminated soil, *Global NEST Journal*, 15(4), 474-484.

[15] Aremu, M. O., Araromi, D. O. and Gbolahan, O.O. (2015).Regeneration of used lubricating engine oil by solvent extraction process. *International Journal of Energy and Environmental Research*, 3(1):1-12

[16] Odokuma, L.O. &Ibor, M.N. (2002). Nitrogen fixing bacteria enhanced bioremediation of crude oil polluted soil. *Global Journal of Pure and Applied Science*, 8(4),455-468.

[17] Nwogu, T.P., Azubuike,C.C. and Ogugbue, C.J. (2015). Enhanced bioremediation of soil artificially contaminated with petroleum hydrocarbons after amendment with *Capra aegagrus hircus* (goat) manure. Biotechnology research international. 2015:22-29

[18] Tsuneo, W. (2002).Pictorial atlas of soil and seed fungi; Morphologies of cultured fungi and key to species.Second edition, CRC press, Boca Raton, London, Newyork. Washington D.C. Pp 1120-1125

[19] Jaqueline, M.C., Paulo, R.M., Renato, N.M., Ivo, S.T., Natalia, M.G.S. and Ederio, D.B. (2013). Toxicity assessment of contaminated soil using seeds as bioindicators. *Journal of Applied Biotechnology*. 1(1):34-40

[20] Millioni, V.S., Servulo, E.L.C., Sobral, L.G.S. and De Carvalho, D.D. (2009). Bioremediation of crude oil-bearing soil: evaluating the effect of Rhamnolipid addition to soil toxicity and to crude oil biodegradation efficiency. Global NEST journal.11(2):181-188

[21] Akpe, A. R., Ekundayo, A. O., Aigere, S.P. and Okwu, G. I.(2015). Bacterial degradation of petroleum hydrocarbons in crude oil polluted soil amended with cassava peels. *American Journal of Research Communication***,** 3(7), 99- 118

[22] Ubalua, A.O. (2011). Bioremediation strategies for oil-polluted marine ecosystems.*Australian Journal of Agricultural Engineering*. 2(6), 160-168.

[23] Agarry, E.S. and Jimoda, L. (2013). Application of carbon-nitrogen supplementation from plant and animal sources in in-situ soil bioremediation of diesel oil: experimental analysis and kinetic modelling. *Journal of Environment and Earth Science*. 3 (7): 51-62

[24] Thapa, B., Ajay, K.C.K. and Ghimere, A. (2012). A review on bioremediation of petroleum hydrocarbon contaminants in soil. *Kathmandu University Journal of Science, Engineering and Technology*. 8(1):164-170

[25] Robertson, G.P. and Groffman, P.M. (2007).Nitrogen transformations. In: Eldor, A. Paul (ed.) *soil microbiology and biochemistry.*3rd edition. Pp 341-347. Elsevier Incorporated. Oxford, United Kingdom.

[26] Okoh, I.O. (2006). Biodegradation alternative in the cleanup of petroleum hydrocarbon pollutants. *Biotechnology* and *Molecular Biology Review*. 1(2):38-50

[27] Teng, Y., Luo, Y., Ping,L., Zou, D, Li, Z. and Christie, P. (2010). Effects of soil amendment with different carbon sources and other factors on the bioremediation of an aged PAH-contaminated soil. *Biodegradation*. 21:167-178

[28] Margesin, R., Hammerle, M. and Tscherko, D.(2007). Microbial activity and community composition during bioremediation of diesel oil-contaminated soil: Effects of hydrocarbon concentration, fertilizers and incubation time. *Microbial Ecology*, 58(2), 259-269

[29] Dadrasnia, A. &Agamuthu, P. (2010).Enhanced degradation of diesel-contaminated soil using organic wastes.*Malaysian Journal of Science*.29, 225-230.

[30] Stephen, E. and Temola, O.E. (2014). Enhanced biodegradation of spent lubricating oil contaminated soil using poultry litter. *British Biotechnology Journal*, 4(8), 868-876

[31] Bento, F.M., Camargo, F.A.O., Okeke, B.C. and Frankenberger, W.T. (2005). Comparative bioremediation of soils contaminated with diesel oil by natural attenuation, biostimulation and bioaugmentation. *Bioresource Technology*. 96(9): 1049-1055

[32] Seklemova, E., Pavlora, A. and Koracheva, K.(2001). Biostimulation-based bioremediation of diesel fuel field demonstration. *Biology*. 12:311-316

[33] Padayachee, D. and Lin, J. (2011). The effect of fertilizer amendment on diesel biodegradation in contaminated soils. *African Journal of Microbiology Research*. 5(14):1729-1739

[34] Stephen, E., Job, O.S. and Abioye, O.P. (2013). Study on biodegradation of diesel-contaminated soil amended with

cowpea chaff. *Journal of Science and Multidisciplinary Research*. 2(1)14-18

[35] Ibiene, A.A., Orji, F.A., Ezidi, C.O. and Ngwobia, C.L. (2011). Bioremediation of hydrocarbon-contaminated soil in the Niger Delta using spent mushroom compost and organic wastes. *Nigerian Journal of Agriculture, Food and Environment*.7(3):1-7

[36] Chaudhry, S., Luhack, J., Sharma, V. and Sharma, C. (2012). Assessment of diesel-degrading potential of fungal isolates from sludge soil of petroleum refinery, Haryana.*Research journal of Microbiology and Biotechnology*. 28:1-9

[37] Chikere, C.B., Okpokwasili, G.C. and Chikere, B.O. (2011). Monitoring of hydrocarbon remediation in soil. *Biotechnology*.1(3):117-138

[38] Okechalu, O.B., Oke, F.M., Egbere, O.J. and Okechalu, J.N. (2014). Effects of biostimulation with organic amendment on the growth and yield of maize (*Zea mays*) in diesel oil-polluted soil. *Journal of Pharmacy And Biological Sciences*. 8:40-44

[39] Agbogidi, O.M. and Ilondu, E.M. (2013). Effects of spent engine oil on the germination and seedling growth of *Moringa olifera* (*Lam.*). *Scholarly Journal of Agricultural Science.* 3(6):293-243

https://dx.doi.org/10.22161/ijeab.51.24 *ISSN: 2456-1878*

Effects of L-arginine on Some Cytogenetical and Physiological Parameters of *Allium cepa* L. Seeds exposed to Salinity

Dilek Çavuşoğlu

Department of Plant and Animal Production, Isparta University of Applied Sciences, Isparta, Turkey.

Abstract— *In this study, L-arginine (Arg) effects on the seedling growth (fresh weight, radicle length and radicle number), seed germination, mitotic activity, chromosomal aberrations and micronucleus frequency in Allium cepa L. germinated in both salt stress and normal conditions were investigated. In only Arg medium, the radicle number of the seeds was partially reduced compared to the control seeds germinated in the distilled water medium, the fresh weight, radicle length and germination percentage indicated statistically the same values as the control. Besides, the mitotic index in the root tip meristems of A. cepa seeds germinated in the Arg medium alone showed a decrease compared to the control seeds germinated in the distilled water medium, whereas the chromosomal aberrations exhibited a significant increase compared to the control. Moreover, the micronucleus formation increased compared to the control. On the other hand, salt stress significantly inhibited the seedling growth and seed germination of A. cepa. In addition, it significantly reduced the mitotic index in the root tip meristems of the seeds and increased the number of chromosomal abnormalities and micronucleus frequency, which is the simplest indicator and the most effective of cytological damage. Nonetheless, the inhibitive effect of salt on the micronucleus formation, mitotic activity, seedling growth, seed germination and chromosomal aberrations significantly decreased with the application of L-arginine.*

Keywords— ***Allium cepa L., arginine, salt stress, seed germination, mitotic index.***

I. INTRODUCTION

Salinity or salt stress is an important abiotic factor that limits crop production. Soils with electric conductivity more than 40mM NaCl (about 4ds/m) are considered to be salty. Approximately 7 % of the earth's land [1] and 20 % of the irrigated land [2] are affected by salinity. More land is irrigated every year to increase crop production. Expansion of irrigated land, coupled with high salt content in irrigation water and poor drainage has increased salinity stress. Blumwald and Grover [3] predict that approximately 50 % of arable land will be affected by salt stress in 2050. Improvement of drainage to improve crop production in saline soils, irrigation management, use and development of salt tolerant varieties are suitable solutions. Salt tolerance is that plants complete their life cycles and grow with good yield potentials in salt conditions. There are two salt tolerance mechanisms; osmotic effect that minimizing salt ingress from root to leaf and ionic effect which minimizes intracellular toxicity due to higher salt concentration [4].

Arginine (Arg), one of the twenty standard amino acids necessary for the formation of proteins and peptides, is also used as a nitrogen storage compound in seeds. In addition, Arg is the precursor of polyamines and nitric oxide, which play an important role in response to various environmental stresses and in many developmental processes. Arg is ideal for storing nitrogen due to its very high N:4 / C:6 ratio, which is a basic amino acid with $H_2NC(=NH)NH(CH_2)_3CH(NH_2)COOH$ linear formula. Indeed, Arg can be represented as a free amino acid in seeds such as soybean, pumkin, broad bean, many other dicotyledonous plants, peach tree or as an important part of the nitrogen store in, other parts of plants or bulbs. And this is an important amino acid in the free amino acid ponds of seedlings of early growth of both seedling growth and megagametophine. Therefore, its biosynthesis may be of additional importance during seed development. Ornithine, is a precursor of Arg and Arg is required for polyamine production. These small positively charged organic molecules play an important role in senescence, root growth, division, response to plant stress, cell growth, ripening, fruit development and other processes. Arg which is the source of nitric oxide plays an important role in root growth, defense, germination, responses, flowering and hormonal marking in plants. Arg is a dietary precursor for nitric oxide formation and nitric oxide is a potent mediator of vascular tone, hence affects the cardiovascular system. The nitric oxide formed from dietary Arg has been linked to muscle repair and

optimal immune function [5]. Feirer [6] stated that the level of Arg increased twenty-fold during the embryo development in the seed. In addition, Arg which is a precursor of polyamines having a regulatory role in embryogenesis. Moreover, it has an important position in a transport compound and the urea cycle too [7].

Since the early 1920s, *Allium cepa* was used to assess the chromosome abnormalities. The method is a sensitive and easy tool to measure total toxicity caused by chemical treatments expressed by growth inhibition of onion bulb roots. *Allium* test was used as a standard test for cyto-genotoxicity monitoring [8]. The test has some advantages, such as being very inexpensive, easy to apply, simple and also as reliable as the method in which abnormalities are recorded in all types of mitotic cells. It combines two test targets: using for toxicity&genotoxicity monitoring, it is also an important fact that it shows a good correlation with mammalian test systems [9].

Although there are few published studies on the role of Arg on the seedling growth and seed germination under both saline and normal conditions, unfortunately, there are no studies on the effects of this amino acid on the micronucleus frequency, mitotic activity and chromosomal aberrations in saline and normal conditions. For these reasons, this work was designed to investigate the effects of Arg in reducing of the harmful effects of salt stress on the mitotic activity, seed germination, chromosomal aberrations, micronucleus frequency and seedling growth of *Allium cepa* L.

II. MATERIALS AND METHODS

2.1. Seed, arginine and salt concentrations

In the present work, *Allium cepa* L. seeds and 0.175 M NaCl (salt) concentration were used. The concentration of L-arginine used in the experiments was 10 mg/L. Arg was obtained from Merck. In a preliminary investigation of this study, Arg and salt concentrations were determined conducted.

2.2. Seed germination

Seed germination experiments were performed in a (fixed temperature) incubator set to 20°C in the dark. Approximately equal-sized and healthy onion seeds have selected. *Allium cepa* L. (*Amaryllidaceae*) seeds have sterilized with the aid of sodium hypochloride solution (2.5%) for ten minute and washed with ultra-pure water for 24 hour. Twenty seeds selected from each application group were placed in plastic containers. The bulbs have split in four groups:

- Group I (control) during 7 sequential days have treated by distilled water.
- Group II during 7 sequential days have treated by 0.175 M NaCl alone.
- Group III during 7 sequential days have treated by a 10 mg/L dose of Arg.
- Group IV during 7 sequential days have treated by a 10 mg/L dose of Arg + 0.175 M NaCl.

It is assumed that the seeds in plastic containers placed in the incubator for germination should have a length of 10 mm. After 7 days, the final germination percentage was taken, the number of radicle were recorded, the radicle lengths of onions were measured in mm, the fresh weights were also determined in g/seed. All experiments were repeated 3 times.

2.3. Cytological and statistical analysis

After a few days for cytogenetic analysis, 1-1.5 cm segment of germinated *A. cepa*'s root tips were excised. Initially, these have pretreated using saturated para-dichlorobenzene for four hours, afterwards were fixed in a solution (3:ethanol / 1: acetic acid) for 24 hours at room temperature and stock up in 70 % ethanol at 4°C until making the microscopic slides. *A. cepa* rootlets were hydrolysed for 15 minutes in 1 N HCl at 60°C, dyed with Feulgen for 1-1.5 hours and lysed with a drop of 45 % CH_3COOH. Squashes have prepared as suggested by Sharma and Gupta [10]. At the end of 24 hours, microscopic preparations were made permanent by means of balsame. With a digital camera (Olympus C-5060) mounted on the Olympus CX41 microscope has photographed mitotic phases, micronuclei and mitotic aberrations (500X).

The cell division densities of these preparations were analyzed by calculating the mitotic index (%) (MI) assessed by analyzing at least 30000 cells per sample (about 10000 per preparation). Chromosomal abnormalities were calculated as the percentage of 2000 dividing cells counted for each concentration. The latter was determined as a percentage between the number of dividing cells (N') and the total number of cells analyzed (N) according to formula: MI (%) = (N' / N) x 100 [11]. The statistical analysis was carried out using SPSS program according to DMRT. Statistically, all values mentioned in this study are highly significant ($P<0.05$).

2.4. Micronucleus (MN) assay

For micronucleus analyses, 1000 cells per slide were scored. MN was examined with the help of a binocular light microscope. For the scoring of micronucleated cells, Fenech et al. [12] used the protocol they followed. These: (i) the diameter of the micronucleus should be a tenth of

https://dx.doi.org/10.22161/ijeab.51.24 ISSN: 2456-1878

the main nucleus, (ii) Micronucleus should be separated from or marginally overlapped from the main nucleus, provided that the nucleus boundary is clearly defined, (iii) the micronucleus staining should be similar to that of the main nucleus.

III. RESULTS

3.1. Effects of arginine on the seedling growth and seed germination

The results from Table 1 clearly demonstrate that while the radicle length, fresh weight and germination percentage of group III germinated in alone Arg medium showed statistically the same values as group I (control) germinated in distilled water medium, their radicle number partly decreased according to the control seeds.

NaCl exhibited an inhibitory effects on all growth parameters examined. For instance, the control (group I) seeds germinated in distilled water medium after 7 days showed 100 % germination, whereas this value was 23 % in group II seeds germinated at 0.175 M salinity. That is to say, NaCl prevented 77 % germination of *Allium cepa* seeds. The inhibitive effect of salt stress on the seed germination was markedly mitigated by Arg application. Group IV seeds treated with Arg in this salt level demonstrated 82 % germination. In addition, Arg continued its success on the seedling growth parameters such as the fresh weight and radicle number. However, this amino acid has been ineffective in attenuating the negative effect of NaCl inhibition on the radicle length. The number of radicle and fresh weight of group II seedlings grown in 0.175 M salinity were 12.7 and 7.0 g, respectively while these values became 18.2 and 11.1 g in group IV seedlings treated with Arg (Table 1, Fig. 1).

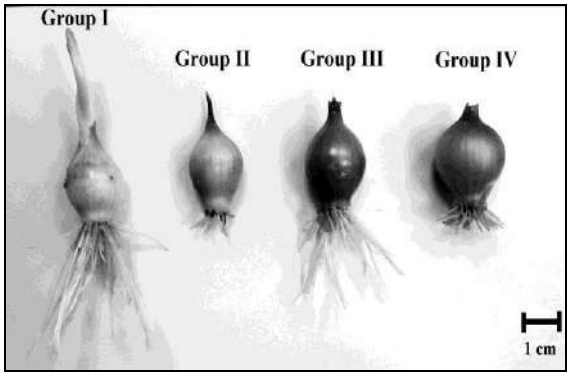


Fig. 1: Allium cepa root tip cells showing germination situations at the end of the seventh day. Control (Group I) seeds were treated with distilled water; Group II seeds were treated with 0.175 M NaCl alone; Group III seeds were treated with a 10 mg L^{-1} dose of L-arginine; Group IV seeds were treated with a 10 mg L^{-1} dose of L-arginine+0.175 M NaCl. Scale bar = 1 cm

Table 1: Effects of L-arginine on some growth parameters of Allium cepa L.

Groups	Growth parameters			
	Germination percentage (%)	Radicle length (mm)	Radicle number	Fresh Weight (g/ seedling)
Group I	*100 ± 0.0^{c}	63.5 ± 0.5^{b}	63.2 ± 0.6^{d}	14.2 ± 0.8^{c}
Group II	23 ± 2.8^{a}	10.3 ± 0.3^{a}	12.7 ± 0.5^{a}	7.0 ± 0.5^{a}
Group III	100 ± 0.0^{c}	63.4 ± 1.0^{b}	56.0 ± 0.5^{c}	14.3 ± 0.3^{c}
Group IV	82 ± 2.8^{b}	11.1 ± 0.9^{a}	18.2 ± 0.5^{b}	11.1 ± 0.6^{b}

*The difference between the values in each column and the same letters isn't significant at the 0.05 level (±SD). Control (Group I) seeds were treated with distilled water; Group II seeds were treated with 0.175 M NaCl alone; Group III seeds were treated with a 10 mg L^{-1} dose of L-arginine; Group IV seeds were treated with a 10 mg L^{-1} dose of L-arginine+0.175 M NaCl.

3.2. Effects of arginine on the micronucleus formation, chromosomal aberrations and mitotic activity

The mitotic index of group III seeds germinated in only Arg medium reduced 20 % compared to group I (control) seeds germinated in distilled water medium. In addition, Arg application increased the chromosomal aberrations and frequency of the micronucleus according to the control (Table 2). Exposure to 0.175 M salinity resulted in a significant inhibition in the mitotic index. In an other words, the mitotic index in the root tip meristems of group II seeds germinated in 0.175 M salt media compared to the group I seeds (distilled water, control) decreased by 89 % and increased significantly the chromosome aberrations (17 %) and the frequency of micronucleus (13 %). On the other hand, Arg treatment became successful in improving

https://dx.doi.org/10.22161/ijeab.51.24 *ISSN: 2456-1878*

the adverse effects of salinity on the micronucleus formation, mitotic activity and chromosomal aberrations. These values became 9.6 % (MN), 9.1 % (MI) and 13.6 % (CAs) in group IV seeds treated with Arg (Table 2).

Figure 2 shows the abnormal mitotic phases observed in course of microscopic examination in meristem cells of *A. cepa* root tip. Double nuclear lesion and micronucleus were the most frequent abnormalities induced by Arg and its salt constituents. Some other aberrations were also observed in cells with the frequency of occurrence as: nucleus disintegration > polar deviation in telophase > anaphase with vagrant chromosome > scattering at metaphase > telophase with vagrant chromosomes > condansed nuclei > uncoiling chromosomes > alignment anaphase > laggards at anaphase > notched nuclei > ring chromosome > nuclear bud > giant cell > prophase with chromosome loss > anaphase with chromosome loss > disturbed in telophase.

Table 2: Effect of L-arginine on some cytogenetic parameters of Allium cepa L.

Groups	Mitotic index (%)	Micronucleus frequency (%)	Chromosome aberration (%)
Group I	*****11.6 ± 1.0^{c}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}
Group II	1.2 ± 0.2^{a}	13.0 ± 1.0^{d}	17.0 ± 0.4^{c}
Group III	9.2 ± 0.5^{b}	1.3 ± 0.5^{b}	14.3 ± 0.5^{b}
Group IV	9.1 ± 0.3^{b}	9.6 ± 0.5^{c}	13.6 ± 1.3^{b}

*The difference between the values in each column and the same letters isn't significant at the 0.05 level (±SD). Control (Group I) seeds were treated with distilled water; Group II seeds were treated with 0.175 M NaCl alone; Group III seeds were treated with a 10 mg L^{-1} dose of L-arginine; Group IV seeds were treated with a 10 mg L^{-1} dose of L-arginine+0.175 M NaCl.

IV. DISCUSSIONS

4.1. Cytogenetical and physiological effects of exogenous arginine under normal conditions

If stress conditions are not present in the environment, any plant growth regulator should be added as exogenous in the germination process. The addition of a plant growth regulator exogenously under stress-free conditions can have negative or positive effect on the seedling growth and seed germination [13, 14]. However, there are few studies on the effects of Arg on the seed germination and seedling growth under normal conditions. Therefore, in the laboratory study, the effects of Arg application on the mitotic activity, seed germination, micronucleus frequency, chromosomal aberrations and seedling growth under normal conditions requested to be tested. The seed germination depending on the used concentration, application method and plant species.

laboratory study's results revealed that the germination percentage, fresh weight and radicle length of the seeds germinated in the only Arg medium statistically showed the same values as the control seeds germinated in distilled water medium, whereas their radicle number decreased slightly compared to the control (Table 1). El-Bassiouny et al. [15] reported that 0.6, 1.25, 2.5 and 5 mM Arg applications increased significantly the fresh weights of wheat seedlings grown in normal conditions. Samia and Rania [16] determinated that 2.5 mM Arg resulted in obvious enhancement in the radicle length and fresh weights of lupine seedlings under normal conditions. These results are not consistent with the present research findings, so Arg may have different effects on the seedling growth and

https://dx.doi.org/10.22161/ijeab.51.24 *ISSN: 2456-1878*

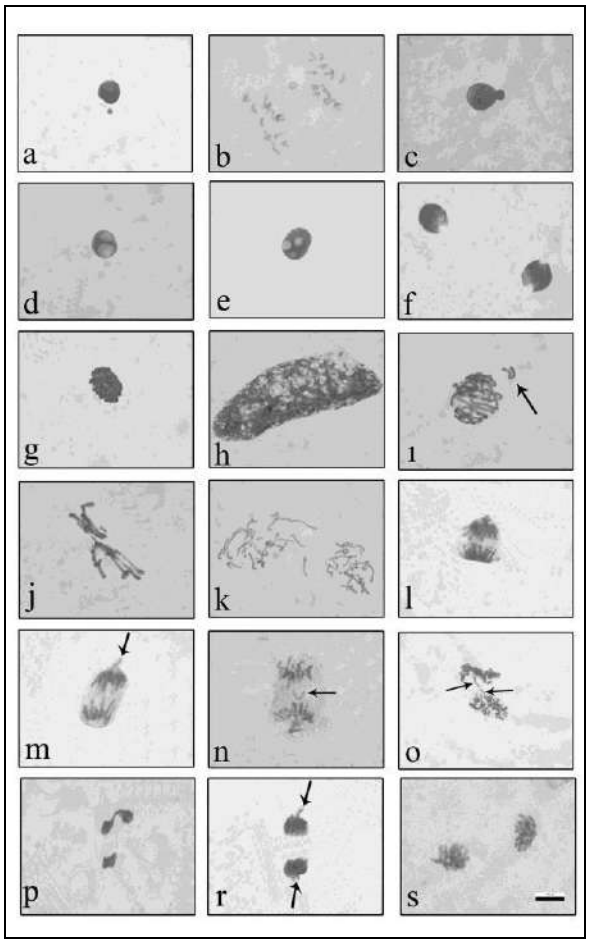


Fig. 2: Chromosomal aberrations; a-micronucleus; b-ring chromosome; c-nuclear bud; d-double nuclear lesion; e-nucleus disintegration; f-notched nuclei; g-condansed nucleus; h-giant cell; ı-prophase with chromosome loss=arrow; j-uncoiling chromosomes; k-scattering at metaphase; l-alignment anaphase; m-anaphase with vagrant chromosome=arrow; n-anaphase with chromosome loss=arrow; o-laggards at anaphase=arrows; p-disturbed in telophase; r-telophase with vagrant chromosomes=arrows; s-polar deviation in telophase. Scale bar = 10 µm

The effects on the chromosomal aberrations, mitotic activity and micronucleus frequency of Arg application under normal conditions are still unknown. The findings of this study showed that the MI in root meristems of *A. cepa* seeds subjected to Arg application under normal conditions decreased about 20 % compared to the control seeds germinated in distilled water medium. Thus, administration of 10 mg L^{-1} Arg demonstrated an effect repressive on the mitotic activity by slowing cell division. This dose of Arg application increased markedly CAs and MN frequency according to the control. In this case, some abnormalities can be said to be caused by this stimulator (Table 2). In this study, micronucleus and double nuclear lesion were the most frequent abnormalities. Various abnormalities have been observed in all stages of mitotic division as a result of structural deviations in chromosomes. A part of the chromosomes can be connected to each other instead of separating to the poles forming fragments and bridges. Laggards observed occured during the chromosomal migration to the poles. Vagrant chromosome in anaphase may also be mainly the result of non-polar mitotic spindles.

4.2. Cytogenetical and physiological effects of exogenous arginine under saline conditions

Salt stress like many other abiotic stresses inhibits plant growth. Because the high salt concentration in the soil solution prevents from being absorbed the nutrient ions in a balanced manner by the plants. NaCl stress affects plant physiology in all plants and also affects cellular levels through osmotic and ionic adjustments resulting in a reduction in biomass production. The negative impact of salt stress is possible to see at all plant levels in almost all growth stages including, seedling, germination, maturity and herbal stages. Although salinity causes ionic and osmotic stress, it causes ionic imbalances that may induce potassium deficiency and may impair the selectivity of root membranes [17]. The results from Table 1 clearly demonstrated that as expected the seedling growth and germination of *A. cepa* seeds were inhibited under saline conditions. In agriculture, soil salinity indicates the presence of high concentrations of soluble salts in the soil moisture of the root zone. Due to their high osmotic pressures, the concentrations of these soluble salts affect plant growth by limiting the water uptake of the roots [17]. Results of these statements are consistent with the results of the present study in terms of showing the decrease in the water content and fresh weight of the seedlings in salted conditions. The inhibitive effect of NaCl on the radicle number and radicle length may result from reducing protein synthesis, nucleic acid and cell division [18].

On the other hand, by application of the amino acid Arg, the inhibitory effect of saltinity stress on the seed germination, fresh weight and radicle number was significantly eliminated (Table 1). To date, there have been several studies investigating the effects of Arg on the seedling growth and seed germination in saline conditions. Abd El-Monem [19] found that the optimum concentration of Arg was 2.5 mM in alleviating the harmful effects of salt stress in wheat. Zeid [20] observed that 4 mM Arg pretreatment promoted the growth parameters and germination percentage of bean seedlings under salinity stress. Nasibi et al. [21] also showed that pre-treatment with three concentrations of Arg (0, 5 and 10µM) could

reduce the harmful effect of salinity on the fresh weight of canola seedlings. Nejadalimoradi et al. [22] observed that 1 and 5 mM Arg pretreatment increased to the radicle length of sunflower plants under salinity stress. In addition, Samia and Rania [16] determinated that spraying 2.5 mM Arg attenuated the retarder effects of salt stress in lupine plants. All of these results are consistent with the amino acid arginine's findings. As can be seen in Table 1, Arg can be understood from the decrease in the osmotic effects of the salt, which relieves the salt stress on the seedling growth and seed germination. For example, in 0.175 M NaCl medium, it is observed that the fresh weight of seedlings is significantly increased by Arg application compared to Group I indicates this probability. Additionally, Arg may have been successful in reducing the inhibitive effects of salinity stress on the seedling growth and seed germination by increasing antioxidant enzyme activities [21].

Mitotic index is a reliable parameter that reflects the frequency of cell division in the root growth area and is used to identify cytotoxicity [8]. Cytotoxicity levels can be determined by a decrease or an increase in the mitotic index [23]. The mitotic index can be used to determine root growth rate and as a reflection of cell proliferation. More interestingly, this study results showed that the salt caused a decrease in the mitotic activity and this decrease was achieved by decreasing the number of cells entering mitotic division. The decrease in the number of divided cells suggests that the salt may have mitodepressive effects on *A. cepa* L. cell division. Mitodepression blocks nucleus proteins and DNA synthesis [24]. With this study, it should be noted that the salinity adversely affects chromosome behaviors and the mitotic activity of *A. cepa* root meristem cells. The results of this study show that salinity decreased MI by 89 % compared to the control group and showed an excessive increase in the number of chromosomal abnormalities and micronucleus. For example, while the MN and CAs in the root tip meristems in group I were 0.0 % and 0.0 %, respectively these values became 13.0 % and 17.0 % in 0.175 M salt. Furthermore, Arg+NaCl became effective in alleviating the harmful effect of salt on the MI. In contrast, administration of simultaneously Arg+NaCl showed a significant success compared to the Arg alone in alleviating the harmful effects of salinity on the frequency of MN and CAs. So, the frequency of CAs with the application of simultaneously Arg+NaCl decreased by 20 %. This result shows Arg repair role against salt injuries during *A. cepa*'s mitosis (Table 2).

Chromosomal abnormalities (CAs), which may occur as a result of both spontaneous and exposure to physical or chemical agents, are characterized by changes in the total chromosome number or chromosomal structure. Chemical and physical agents can induce CAs, which is carried out by different mechanisms including aneugenic and clastogenic actions. While aneugenic effect involves inactivation of a cell structure such as mitotic spindles leading to chromosomal losses, clatogenic effect is characterized by induction of chromosomal break during cell division. Nuclear abnormalities are derived from various types of CAs such as micronucleus, lobed nuclei, mini cells, nuclear buds and polynuclear cells. A number of chromosomal abnormalities are derived from nuclear abnormalities such as micronuclei, lobated nuclei, polinucleated cells, nuclear buds and mini cells. Micronucleus (Fig. 2a) may originate from all chromosomes (clastogenic agent) or from acentric fragments (aneugenic agent) not included in the main nucleus during the cell cycle [25]. Ring chromosomes (Fig. 2b) are the result of chromosome losses in the telomere domain [26]. Nuclear buds (Fig. 2c) associated with the formation of micronucleus are indicative of the initial process to discard nuclear material [23]. According to Akaneme and Iyioke [27], the presence of nuclear lesions (Fig. 2d) indicates cytological evidences for the inhibitory effect on DNA biosynthesis. Giant cells (Fig. 2h) occur due to incomplete cytoplasmic division but they grow up with nuclear division and DNA replication before they die [28]. The presence of vagrant chromosomes (Fig. m, r) means a deviation of mitotic spindle irregularity, an aberration which may result in delayed metaphase and /or prophase [29].

V. CONCLUSION

There are no literature data on the cytogenetic parameters examined in normal and saline conditions. Therefore, the results of this study have been particularly reported for the first time in normal and saline conditions. As a conclusion, this study showed that Arg can significantly increase activations such as the seed germination, seedling growth, mitotic index, MN and CAs under saline or normal conditions.

REFERENCES

[1] Aoki, A., Kanegami, A., Mihara, M., Kojima, T., Shiraiwa, M., Takahara, H. (2005). Molecular characterization of a novel soybean gene encoding a leucine-zipper-like protein induced to salt stress. *Gene*. 356, 135-145.

[2] Flowers, T.J., Yeo, A.R. (1995). Breeding for salinity resistance in crop plants. Where next?. *Aust*ralian *Journal of Plant Physiology*. 22, 875-884.

[3] E. Blumwald and A. Grover, "Salt tolerance: Current and future uses of genetically modified crops" in Plant

Biotechnology, G. Nigel and Halford, Eds. Cambridge: United Kingdom, 2006, pp. 206-224.

[4] Stoddard, F.L., Balko, C., Erskine, W., Khan, H.R., Link, W., Sarker, A. (2006). Screening techniques and sources of resistance to abiotic stresses in cool-season food legumes. *Euphytica*. 147, 167-186.

[5] Garcia, A. G. M. (2017). Hemp: a composition review plus. Senior project for Bachelor of Science Nutrition at California Polytechnic State University San Luis Obispo, C.A., pp. 1-83.

[6] R.P. Feirer, "The biochemistry of conifer embryo development: amino acids, polyamines and storage proteins" in Somatic embryogenesis in woody plants, S.M. Jain, P.K. Gupta, R.J. Newton, Eds. Dordrecht: Kluwer Academic Publishers, 1995, vol. 1, 317-336.

[7] Kärkönen, A. (2001). Plant tissue cultures as models for tree physiology: Somatic embryogenesis of *Tilia cordata* and lignin biosynthesis in *Picea* abies suspension cultures as care studies, *Dissertation*, Helsinki: Univ. of Helsinki.

[8] Fiskesjo, G. (1985). The *Allium* test as a standard in environmental monitoring. *Hereditas*. 102, 99-112.

[9] H. L. Yi and Z. Q. Meng, "Municipal landfill lechate induces cytogenetic damage in root tips of *Hordeum vulgare*" in Acta Phyto Ecotoxicology and Environmental Safety. N. Sang, G. Li, X. Xin, Eds. vol. 63, 2002, pp. 469-473.

[10] Sharma, P.C., Gupta, P.K. (1982). Karyotypes in some pulse crops. *Nucleus*. 25, 181-185.

[11] Aslantürk, Ö.S., Çelik, T.A. (2005). Preventive effect of lycopene on chromosome aberrations in *Allium cepa*. *Pakistan Journal of Biological Science*. 8, 482-486.

[12] Fenech, M., Chang, W.P., Kirsch-Volders, M., Holland, N., Bonassi, S., Zeiger, E. (2003). HUMN (human micronucleus) project: detailed description of the scoring criteria for the cytokinesis-block micronucleus assay using isolated human lymphocyte cultures. *Mutation Research*. 534, 65-75.

[13] Çavuşoğlu, D. (2019). Physiological and cytogenetical effects of L-Tyrosine in *Allium cepa* L. exposed to NaCl stress. *Fresenius Environmental Bulletin*. 28 (12A), 9753-9759.

[14] Çavuşoğlu, K., Doğu, F., Çavuşoğlu, D. (2019). Effects of Sodium Hypochlorite (NaClO) on some Physiological and Cytogenetical Parameters in *Allium cepa* L. Exposed to Salt Stress. *Bangladesh Journal of Botany*. 48(2), 223-229.

[15] El-Bassiouny, H.M.S., Mostafa, H.A., El–Khawas, S.A., Hassanein, R.A., Khalil, S.I., Abd El–Monem, A.A. (2008). Physiological responses of wheat plant to foliar treatments with arginine or putrescine. *Australian Journal of Basic Applied Sciences*. 2(4), 1390-1403.

[16] Samia, A.A., Rania, S.H. (2018). Alleviation of oxidative effects of salt stress in White lupine (*Lupinus termis* L.) plants by foliar treatment with L-Arginine. *Journal of Animal and Plant Sciences*. 28(1), 165-176.

[17] Nawaz, K., Hussain, K., Majeed, A., Khan, F., Afghan, S., and Ali, K. (2010). Fatality of salt stress to plants: Morphological, physiological and biochemical aspects. *African Journal Biotechnology*. 9(34), 5475-5480.

[18] Mccue, K.F., Hanson, A.D. (1990). Drought and salt tolerance: towards understanding and application. *Trends Biotechnol*ogy. 8, 358-362.

[19] Abd El-Monem, A.A. (2007). Polyamines as modulators of wheat growth, metabolism and reproductive development under high temperature stress, *Dissertation*, Cairo: Ain Shams Univ.

[20] Zeid, I. M. (2009). Trehalose as osmoprotectant for maize under salinity-induced stress. *Research Journal of Agriculture and Biological Sciences.* 5, 613-622.

[21] Nasibi, F., Manouchehri, K.K.H., Barand, A. (2014). Effects of seed pre-treatment with L-arginine on improvement of seedling growth and alleviation of oxidative damage in canola plants subjected to salt stress. *International Journal of Plant Production*. 5(1), 1217-1224.

[22] Nejadalımoradı, H., Nasibi, F., Manoochehri, K., Kalantari, K., Zanganeh, R. (2014). Effect of seed priming with L-arginine and sodium nitroprusside on some physiological parameters and antioxidant enzymes of sunflower plants exposed to salt stress. *Agricultural Communications.* 2(1), 23-30.

[23] Fernandes, T.C.C., Mazzeo, D.E.C., Marin-Morales, M.A. (2007). Mechanism of micronuclei formation in polyploidizated cells of *Allium cepa* exposed to trifluralin herbicide. *Pesticide Biochemstry and Physiology*. 88, 252-259.

[24] Schulze, E., Kirschner, S. (1986). Microtubule dynamics in interphase cells. *Journal of Cell Biology*. 102, 1020-1031.

[25] Fenech, M. (2000). The in vitro micronucleus technique. *Mutation Research*. 455, 81-95.

[26] Khanna, N., Sharma, S. (2013). *Allium cepa* root chromosomal aberration assay. *Indian Journal of Pharmaceutical and Biological Research*. 1, 105-119.

[27] Akaneme, F.I., Amaefule, C.C. (2012). Evaluation of the cytotoxicity and genotoxicity of aqueous leaf extracts of *Azardirachta indica* A. Juss. using the *Allium* test. *Journal of Medicinal Plants Research*. 6, 3898-3907.

[28] Prajitha, V., Thoppil, J.E. (2014). Induction of giant cells by the synthetic food colorants viz. lemon yellow & orange red. *Cytotech*. 68(3), 443-450.

[29] Tkalec, M., Malaric, K., Pavlica, M., Pevalek-Kozlina, B., Vidakovic-Cifrek, Z. (2009). Effects of radiofrequency electromagnetic fields on seed germination and root meristematic cells of *Allium cepa* L. *Mutation Research-Fundamental and Molecular Mechanisms of Mutagenesis*. 672, 76-81.

https://dx.doi.org/10.22161/ijeab.51.25 *ISSN: 2456-1878*

Strategy for Development of Rice Sawah Culture Planting in Jarwo Plants with Various Modification of Plant Distance

Utami Paulina, Auzar Syarif, Aswaldi Anwar

Department of Agronomi, Faculty of Agriculture, Andalas University, Padang – West Sumatera, Indonesia

Abstract— *Rice is the main food crop of the Indonesian population, mostly planted in paddy fields. The speed of population growth is not balanced with a decrease in rice production that occurs due to a decrease in harvested area. This research aims to determine the interaction between distance planting in the jarwo planting system on the growth and yields of lowland rice (oryza sativa). This research was conducted in December 2018 until March 2019 in Andalas Makmur, Parak Karakah, Kuranji District, Padang. The method used was a Randomized Block Design (RBD) with 2 factors governing spacing, consisting of 3 levels, namely J1: Distance planting (20 x 25) cm², J2: Planting distance (25x 25) cm², J3: Planting distance (30 x 25) cm², The second factor of the jarwo planting system consists of 4 levels, namely L1: Jajarlegowo 2: 1, L2 : Jajarlegowo 3: 1 A, L3: Jajarlegowo 3: 1 B, L4: Jajarlegowo 4: 1. In this experiment there were 12 combinations. Each combination consists of 3 groups. The observational data were statistically analyzed by the F test and if the F test count was greater than the F table of 5%, then continued with Duncan's New Multiple Range Test (DNMRT) at the 5% significance level. The results showed that there was no interaction between planting distances in various jarwo planting systems on growth and yields of paddy (oryza sativa).*

Keywords— ***Rice, Spacing, Jajarlegowo.***

I. INTRODUCTION

Rice is the main food crop of the Indonesian population, mostly planted in paddy fields. The speed of population growth is not balanced with a decrease in rice production that occurs due to a decrease in harvested area. Indonesia's food security, independence and sovereignty are not yet considered sturdy. This is indicated by high imports of food products. Until 2013 the problem of food security, especially rice, became a major problem for the Indonesian people. In 2011, imports were 1.6 million tons and in 2012 rice imports were 1.9 million tons (Pujiasmanto, 2013). In addition, the low productivity of rice is caused by the pattern of planting and harvesting of inter-regional wetland rice that is not uniform, agricultural technology innovation is still low, and land use is not yet optimal. The main problem of rice in cultivation is its stagnant productivity. In the past decade, the increase in rice yield per ha was not significant. Nationally, in 2011, productivity only reached around 5 tons per ha. On the other hand, the availability of paddy fields is also difficult to develop. Therefore, it is necessary to continue to look for cultivation methods that can increase yields and productivity.

The fundamental problem of food agriculture (rice) and is classic in Indonesia is narrow land, an average of 0.2-0.3 ha per farmer family. In the long history of agriculture, narrow land is not able to make farmers achieve economic and welfare levels. Indeed the government program has been able to increase rice production, but it is not always accompanied by improvements in farmers' welfare. One key to increasing production is through increasing productivity, while productivity cannot be separated from the role of agricultural technology.

As a staple food, rice cannot be replaced with other carbohydrate sources. That is because of the Indonesian people's eating habits, if you haven't eaten rice then it means you haven't eaten. Indonesian people's rice consumption based on the Central Statistics Agency in 2017 shows 1,571 / kg / week and 2017 rice production is 81.3 million tons / year (BPS, 2018). Over the past 37 years the average annual rice consumption is higher than the average annual rice production, therefore domestic rice production often does not cover domestic rice consumption (Kusmanaet., Al, 2017).

https://dx.doi.org/10.22161/ijeab.51.25 *ISSN: 2456-1878*

According to Karokaro*et al*., (2014) in an effort to achieve the government's target of increasing national rice production (P2BN) in this case the Ministry of Agriculture through development and research agencies has issued many recommendations to be applied by farmers. One of these recommendations is the application of a correct and good planting system through spacing of planting known as the jajarlegowo planting system. In principle, the jajarlegowo planting system is to increase the population by adjusting the spacing. This planting system also manipulates the layout of the plants, so that most of the clumps of plants become edge plants (Ikhwani et al. 2013).

Jarwo planting system is structuring rice plants by adjusting the spacing in such a way as to achieve optimal plant population and the number of plants that get more side effects than the usual planting method, so as to produce higher productivity than conventional planting methods. This has been proven by farmers who apply well, rice farming with the Jarwo planting system on average is able to increase rice productivity.

Some obstacles in applying the Jarwo planting system are as follows: The application of the Jarwo planting system is not correct, which is seen from the lower number of plant populations compared to conventional systems / tiles so that productivity is not significantly different or lower than conventional planting methods, planting power is scarce, while in the other side of the Jarwo system requires more expensive planting costs, there is a slash system where traders value the yield of production per ha is not different between the Jarwo system and the tiled system so that farmers do not get incentives to implement the Jarwo planting system, and planting a single seed in the Jarwo planting system also farmers are still doubtful because farmers are accustomed to planting 2-3 seeds per hole.

Furthermore according to (Santoso et al., 2005) several factors that cause the development of legowo row planting are not: (1) a wholesale planting system that requires faster planting time, while the legowo planting system requires a longer time; (2) the limited number of planting workers skilled in the application of the Legowo planting method, and (3) the higher the Legowo planting costs.

But what we need to know about the use of plant spacing is basically to give the possibility of plants to grow well without experiencing much competition in terms of taking water, nutrients, and sunlight. Proper spacing is important in optimizing the use of sunlight for photosynthesis. In the right planting distance, plants will get a balanced growth space. The effect of the rice planting system as a component of cultivation that has an effect on yield and income, is apparently complex (Makarim et al. 2005). Plant spacing and plant orientation in the field affect the following six important processes: (1) capturing solar radiation by plants for photosynthesis, (2) absorption of nutrients by the roots, (3) plant water requirements, (4) circulation of photosynthetic CO2 and O2 resulting from photosynthesis, (5) availability of space that determines weed populations, and (6) microclimate under the canopy, which influences the development of plant pests (OPT). The results of spacing in Indonesia reported Pratiwi et al. (2010) concluded that wide spacing provides opportunities for plant varieties to express their growth potential. The denser the plant population, the smaller the number of tillers and the number of panicles per clump. In low populations (wide spacing), the performance of large rice groves, but the breadth of yield and yield components is lower than denser spacing.

The right spacing will give maximum growth, number of tillers, and yields. According to Sohel et al. (2009), the optimum spacing will provide good growth of the top of the plant so that it can utilize more sunlight and the growth of the roots which is also good so that it can utilize more nutrients. Conversely, planting spacing that is too tight will result in very intense competition between plants in terms of sunlight, water, and nutrients. As a result, plant growth is inhibited and crop yields are low.

To overcome these obstacles, the Government is making efforts to re-highlight the way rice cultivation is highlighted and appointed as one of the breakthroughs in increasing rice productivity, namely the jajarlegowo planting system. Legowo row rice planting system is one of the cultivation techniques that can provide opportunities for rice plants to provide various facilities including ease in application of fertilizer, weed control and control of plant pests.

Orientation of legowo row cropping although in the same population has the opportunity to produce higher grain because of the more photosynthesis that occurs, because it is more effective in capturing solar radiation and the easy diffusion of CO2 gas for photosynthesis. Lin et al. (2009), states that wide spacing can improve total light capture by plants and can increase seed yield. Greater distance between rows can improve the total light radiation captured by plants and can increase yields.

Therefore, the application of the Legowo row planting system in accordance with local environmental conditions will almost certainly increase rice productivity and profits for farmers, while national expansion can increase rice production. Legowo planting system 2: 1 or 3: 1 and 4: 1, is

https://dx.doi.org/10.22161/ijeab.51.25 *ISSN: 2456-1878*

an alternative technology component in irrigated rice. The choice of Integrated Plant Management (PTT) technology component is based on the identification of the area and the problems of rice farming which are expected to be an opportunity to overcome the problem of doubling rice productivity (Basri, et. Al. 2010). Peripheral plants grow and develop better and yields per clump are higher than those in the middle, so that more and more border effect edge crops in rice fields produce more grain. For that reason, in terms of increasing rice production, it is highly recommended to use the Legowo row planting system. Because, the components of rice yield (apart from being determined by the type of variety and yield level) are significantly affected by spacing, especially the amount of grain and panicle length (Pratiwi et al., 2009). This spacing is regulated not only to regulate plant neatness but also to be used as populations or clumps, so as to overcome the problem of rice productivity, it is necessary to have a new technology and innovation in agricultural production, namely by using new spacing patterns, namely the legowo row system in rice cultivation.

II. RESEARCH METHODS

A. Time and Place This research took the form of a field trial, which was carried out from December 2018 to March 2019 in Andalas Makmur, ParakKarakah, Kuranji District, Padang. The schedule of activities can be seen in Appendix 1.

B. Materials and Tools

The materials used in the research are IR 42 Varieties of Rice Seeds (description can be seen in Appendix 2), raffia ropes, manure, NPK 15 inorganic fertilizers, Urea, and insecticides for pest control (darmabas and ribcorde). The tools used in this experiment were plow, machete, rake, seed bed, hoe, meter, calipers, scales, scythe, 15 L sprayer and stationery.

C. Research Design

This research was in the form of a factorial 2-factor experiment that was designed according to a randomized group (RBD). The first factor is spacing which consists of 3 levels, namely:

PP1: Spacing (20 x 25) cm²
PP 2: Spacing (25x25) cm²
PP3: Spacing (30 x 25) cm²

The second factor is the jarwo planting system which consists of 4 levels, namely:

L1: Jajarlegowo 2: 1
L2: Jajarlegowo 3: 1 A
L3: Jajarlegowo 3: 1 B
L4: Jajarlegowo 4: 1

The layout of the experimental plot is in Appendix 5 and the plant population and samples in each experiment per plot are in Appendix 4. In this experiment there are 12 treatment combinations. Each combination consists of 3 groups so that there are 36 total experimental units. The observational data were statistically analyzed by the F test and if the F test count was greater than the F table of 5%, then continued with Duncan's New Multiple Range Test (DNMRT) at the 5% significance level.

III. RESEARCH IMPLEMENTATION

A. Land Preparation

Land preparation begins with plowing twice, the first is to use a tractor by turning the soil over and flooded during processing. The second hijacking is done a week after the first hijacking, and when the second hijacking is done at the same time as the ground smearing. A few days after puddling the soil was leveled and mapped 36 plots of 3 x 4 m in size and 0.75 m in spacing between groups in 1 m.

B. Nursery

The nursery is carried out in the experimental field with 2x 1m seedling size. The varieties planted in this study are IR 42. Preparation for the nursery is carried out; (1) Rice seed selection is by inserting rice seeds into a bucket that has been filled with water, (2) Floating seeds are discarded and seeds that are immersed are soaked for 2x24 hours and air dried for 12 hours until they germinate. (3) Germinated seeds are sown at the nursery for 21 days.

C. Labeling

Labeling is done after making the test plot according to the treatment, the label is made of plastic label.

D. Planting

The age of the seeds of rice plants used is 21 days after seedling (HSS). Planting was carried out simultaneously for all treatments with 3 stems per planting hole. Spacing and planting systems are adjusted according to the treatment. The distance of the plants is 20x25 cm, 25x25 cm and 30x25 cm with each jarwo different types, namely 2: 1, 3: 1A, 3: 1B, and 4: 1.

https://dx.doi.org/10.22161/ijeab.51.25 *ISSN: 2456-1878*

E. Maintenance

Maintenance carried out include:

E.a. Stitching

This stitching is done by using seeds planted outside the ranks of the Legowo ranks. These seeds are planted simultaneously with planting in the fields. Planted seedlings are given a distance of 10 cm from plants that have been planted. It is intended that if the main seedlings in the row die because the snail pest is kept then the seedlings will be replaced by enlarging into the ranks. The seeds used for replanting come from early peace. Replanting is done no later than 7 days after planting

E.b. Weeding weeds

Weed weeding is done a week after planting until the end of the vegetative period with a interval of once a week. Weeding is done when weed populations grow in the study area according to conditions in the field. Weed weeding is done manually by pulling weeds in the study area, which was flooded first to make it easier to weed.

E.c. Fertilization

Fertilization is done by sowing. Fertilization of empty rows between 2 rows of plants. Fertilizer is sown to the left and right evenly, so that in one go can fertilize two rows of plants. Fertilization is done using Urea Fertilizer with a recommendation of 250 kg / ha, which is done 2 weeks after planting and 5 weeks after planting, SP 36 100kg / ha which is done at the age of 5 weeks after planting and andKCl 100kg / ha is done at 5 weeks after planting (Appendix 3).

F. Harvesting

Harvesting is done when the rice plants have yellowed more than 90% in one plant family and the leaves have dried, namely when the rice plant is 18 MST. Harvesting is done by mowing the rice plants using a sickle, after which threshing is done to separate the rice grain.

A. Plant height (cm)

Observations were made by measuring plants from the base of the stem (root neck) to the longest leaves in the sample plants using a meter. Observation starts 2 weeks after planting to 8 weeks after planting with an observation interval of 2 weeks.

B. Total Number of Puppies (clumps)

Observation of the number of tillers per clump in the sample plants began 2 weeks after planting to 8 weeks after planting with an observation interval of 2 weeks. The number of tillers was calculated by counting all the stems per plant.

IV. RESULTS AND DISCUSSION

A. Plant height (cm)

The height of rice plants at the age of 8 MST did not have a significant effect and there was no interaction between the treatment of spacing and jarwo planting patterns (Appendix 7.D). Rice plant height data can be seen in table 4:

Table 4. The height of a jarwo paddy cropping plant with various spacing and jarwo planting system:

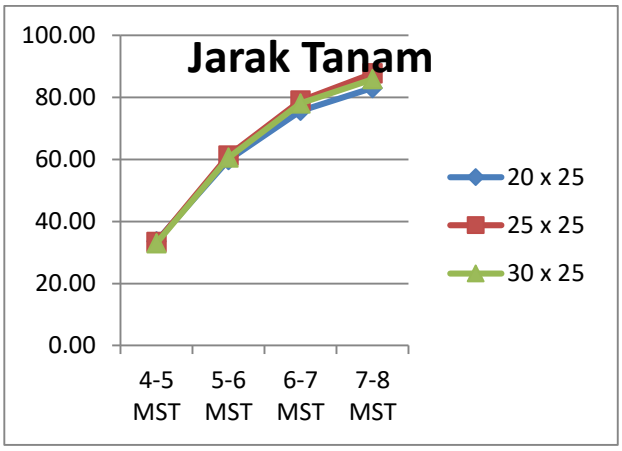
JarakTanam (cm)	JajarLegowa				Rata – rata
	2 : 1	3 : 1A	3 : 1B	4 : 1	
	cm	cm	cm	cm	cm
20 x 25	89,27	89,67	84,20	69,00	87,71
25 x 25	86,73	93,07	89,60	81,53	89,80
30 x 25	84,73	84,00	82,87	91,80	83,87
Rata – rata	86,91	88,91	85,55	80,77	
KK : 10,12%					

Note: the figures on the lane and are not significantly different according to the F test at 5% significance level.

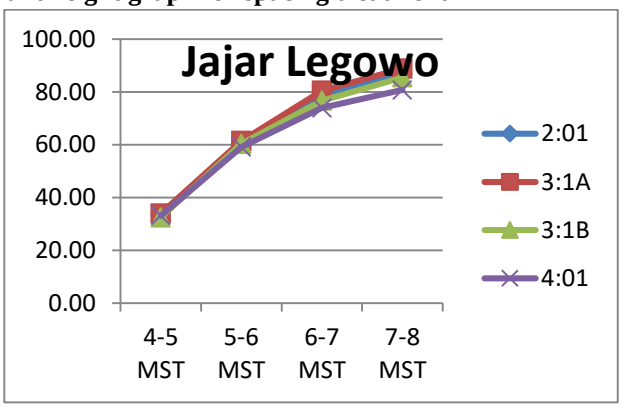
Based on the results of the calculation of variance shows that the treatment of the use of spacing in the planting system jajarlegowo pattern is not significantly different and there is no interaction between the two factors. This is seen from a distance. This is because the jajarlegowo planting system provides wider space so that competition does not occur between plants to get a greater supply of nutrients so that it affects growth and production.

Spacing that is not too tight causes sunlight to enter the planting area which can then be used by plants for photosynthesis. The more plants absorb sunlight will accelerate the process of photosynthesis, as well as the formation of photosynthates so that the filling of grain will be optimal (Supriyanto et al., 2010).

https://dx.doi.org/10.22161/ijeab.51.25 *ISSN: 2456-1878*



Plant height graph for spacing treatment



Plant height chart in the treatment of Legowo row

Fig.4: Development of plant height in jarwo wetland rice cultivation with various modifications of spacing

Based on Figure 4 on the plant height chart we can see that the value of development every week on the height of rice plants in the distance plant treatment is increasing every week as well as in the treatment of the Legowo row type where the increase occurs every week.

In accordance with the opinion of Aribawa (2012), it states that a higher plant height is produced in more plant populations in one stretch. High plant growth does not guarantee crop productivity is also high. Plants that grow well are able to absorb nutrients in large quantities, the availability of nutrients in the soil affects the activity of plants including photosynthetic activity, so that the plant can increase growth and production. The more the distance of planting is used, the higher plant growth will be faster because the plants are trying to find more sunlight (Nursanti, 2009).

Here can be seen in table 4, namely jarwo 4: 1 cropping pattern with a spacing of 30x25 cm which is the lowest average value, This is because the growth of rice plants is more influenced by population density. At high densities competition will occur against sunlight, oxygen, nutrients and water. According to Bozorjai (2011) where the high and low rice plants are strongly influenced by the level of plant density.

Muyasir (2012) states that the increase in plant height is caused by the plant's canopy which is getting closer together resulting in the quality of the light received being decreased. The closer plant spacing is used, the higher plant growth will be faster because the plants try to look for more sunlight. In the treatment of plant spacing, rice plants are not able to develop more rapidly due to high plant density. The ability of seeds to obtain nutrients still experiences competition with the use of tight spacing. According to Mulyaningsihet. al (2008). Competition is a form of relationship between two or more individuals having a negative influence on both parties. Because basically the use of the legowo network system is an effort to increase rice production / yield by manipulating the environment, so as if all plants are peripheral plants, so that the utilization of sunlight can be optimized for photosynthesis. This is in accordance with the opinion of Ikhwani, Gagad, Eman and Makarim (2013), the use of plant spacing is basically giving the possibility of plants to grow well without experiencing much competition in terms of fetching water, nutrients, and sunlight. The right planting distance is important in the optimal utilization of sunlight for photosynthesis. In the right planting distance, plants will get a balanced growth space.

B. Total Number of Puppies (sticks)

The total number of tillers at the age of 8 MST showed no significant influence and interaction between treatment of plant spacing and jarwo system planting patterns (Appendix 7.E). Data on the total number of tillers is shown in table 5.

Table 5. Total number of tillers of jarwo planting patterns with various spacing and jarwo planting systems:

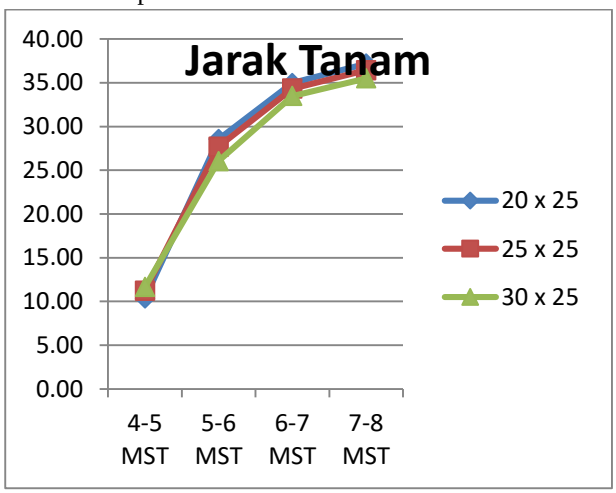
JarakTanam (cm)	JajarLegowa 2 : 1	3 : 1A	3 : 1B	4 : 1	Rata – rata
20 x 25	39,27	38,67	35,73	35,07	37,89
25 x 25	34,73	39,60	36,87	34,53	37,07
30 x 25	36,07	33,53	35,53	36,87	35,04
Rata – rata	36,69	37,26	36,04	35,49	
KK : 10,78%					

Note: the figures on the lane and are not significantly different according to the F test at 5% significance level.

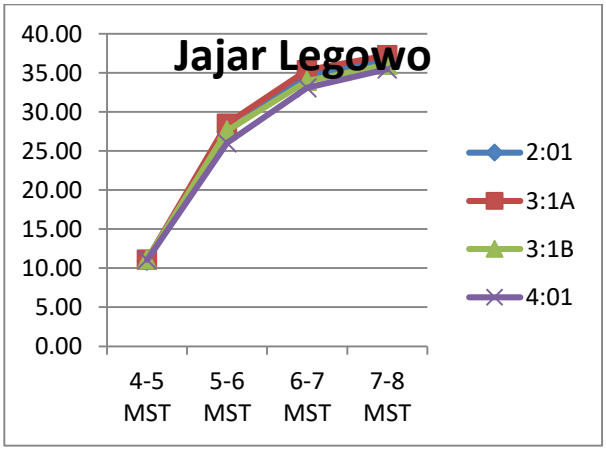
https://dx.doi.org/10.22161/ijeab.51.25 *ISSN: 2456-1878*

Based on the results of the calculation of variance shows that the treatment of the use of spacing with several planting systems jarwo pattern is not significantly different. This is seen in the table we see that there is no interaction between the two treatment factors. This is expected because the number of plant rows and plant populations are not too large and dense, so that the optimal environmental conditions can be used when growing.

Sauki et al. (2014), the maximum number of tillers will affect the number of productive tillers and correlate to the results.



Graph of total number of tillers in spacing



Graph of total number of tillers in spacing

Fig.5. The development of the total number of tillers in jarwo paddy rice cultivation with various modifications to planting spacing

Based on Figure 5 in the graph the total number of tillers we can see that the value of development every week the number of tillers in rice plant spacing increases every week as well as in the treatment of Legowo row type where an increase occurs every week.

This is related according to Husana (2010), the number of tillers will be maximal if the plant has good genetic traits added with favorable environmental conditions or in accordance with plant growth and development.

Furthermore, it was stated that the maximum number of tillers was also determined by spacing, because spacing determined solar radiation, mineral nutrients and cultivation of the plant itself. But genetic and environmental factors also determine the productivity of rice.

Proper spacing is important in optimizing the use of sunlight for photosynthesis, to obtain a balanced growth space. With the occurrence of competition, especially water and unclean elements will disrupt plant growth. The application of the jarwo planting system is able to assist plants in photosynthesis optimally, the existence of an empty space in the form of an aisle that extends the legowo planting system will increase the interception of light and co2 into the crop then it will also increase plant metabolism and biosistesis so that the production of rice plants is more optimal. Increasing the number of rice seedlings every week occurs because young seedlings have better adaptability compared to old seedlings so plants can grow better. Age of seedlings in the field moved very influential on rice production. The faster the field seedlings move, the more adequate the period for seedlings to adapt to the new environment, so the more adequate period for seedling and root development. In addition, the number of rice tillers is also related to the phyllochron formation period. Phyllochron is the period in which a set of stems, leaves and roots emerge from the base of the plant and the subsequent germination. The older the seedlings are moved to the field, the fewer the number of phyllochron produced, while the younger the seedlings are transferred, the more the number of phyllochron produced so that the number of tillers can be produced (Sunadi, 2008).

Unlike the 4: 1 jarwo planting pattern with a spacing of 30x25 cm which results in the least number of tillers. This is suspected because the legowo row planting system is a manipulation of the layout of a plant so that the plantations will have a higher number of edge plants in the presence of an empty row. Although the use of a number of seedlings per planting hole will not have an effect because population densities still occur, so they are not optimal in receiving sunlight.

Besides the influence of population density on the planting system, the formation of tillers is also influenced by genetic traits and environmental conditions that are in accordance with plant growth. According to Asfaruddin (1997) tall plants

https://dx.doi.org/10.22161/ijeab.51.25 *ISSN: 2456-1878*

use more asimilatnya for the formation of stems and leaves than for the formation of tillers.

A wider spacing allows each plant to get more resources so that plant growth and productivity are better. According to Uphoff et al., (2002) that the number of tillers will be maximal if soil fertility and growth space are optimal. The wider spacing between lines in the Legowo range makes it easier to regulate water and control pests and plant diseases and maximize utilization of fertilization. The number of tillers in rice plants does not increase with age as the plant enters the generative phase. The more total number of tillers produced will potentially produce the number of productive tillers.

V. CONCLUSIONS AND RECOMMENDATIONS

Conclusions

There is no interaction between plant spacing in various jarwo planting systems on the growth and yield of lowland rice

Suggestions

Based on the results of the study and conclusions obtained, it is recommended that when conducting research on plant spacing and jarwo cropping system, use the right spacing is not too tight and not too tenuous so that the use of spacing gives the possibility of plants to grow well without experiencing much competition, besides that in the jarwo planting system use the appropriate method of planting the system so that plant growth will be optimal.

REFERENCES

[1] Aribawa, 2012.Effect of cropping systems on increasing rice productivity in wetlands upland wetlands.Bali Institute of Agricultural Technology Assessment (BPTP).Denpasar.Http:// universitas.trunojoyo.ac.id.

[2] Asfaruddin, 1997.Evaluation of upland rice galenggalur tolerability of aluminum poisoning and its efficiency in the use of potassium. Thesis. IPB graduate program. Bogor. Central Bureau of Statistics .2018.Rice Production in Entire Provinces. http://bps.tnmnpgn.go.id. Accessed July 2, 2018.

[3] Basri, F. and Haris M., 2010. Fundamentals of International Economics: Introduction and Application of Quantitative Methods. Kencana: Jakarta.

[4] Husana, Y. 2010. Effect of Use of Spacing on Rice Growth and Production. Thesis. Agriculture Science Study Program Post Graduate Program.Tadulako University. Ikhwani, Pratiwi, Paturrahman, and Makarim, (2013) 'Increased rice productivity through the application of spacing jajarlegowo', Food Crops Science, 8 (2): 72–79.

[5] Karokaro, S., J. E. Rogi, D. S. Runtunuwu, and P. Tumewu. 2014. Spacing of Rice Paddy Planting (Oryza sativa L.) in LegowoJajar Planting System. [Online].Www.portalgaruda.com.Accessed January 23, 2017.

[6] Kusmana A, Budiman A, Hidayat A. 2017. Development of Food Production and Consumption in Indonesia.https: //mpra.ub.unimuenchen.de. Accessed on 2 July 2018.

[7] Lin, XQ, D.F. Zhu, H.Z. Chen, and Y.P. Zhang 2009.Effects of plant density and nitrogen application rates on grain yield and nitrogen uptake of super hybrid rice. Rice Science 16 (2): 138-142.

[8] Makarim, A.K., D. Pasaribu, Z. Zaini, and I. Las. 2005. Analysis and synthesis of the development of integrated crop management models for lowland rice. Indonesian Rice Research Institute. 18p.

[9] Mulyaningsih, 2008.Comparative Analysis of Income of SRI (System of Rice Intensification) with the Conventional System.Faculty of Agriculture.University of Lampung.

[10] Muyassir 2012.Effect of Spacing, Age and Number of Seedlings on Yields of Rice (Oryza sativa L.). J. Land Resource Management. 1 (2): 207-212.

[11] Nursanti, R. 2009. Effect of Seed Age and Planting Distance on Growth and Productivity of Hotong (Setaria italic (L.)Beauv) Crops. Journal of Plant Production.5 (2): 52:54.

[12] Pratiwi, G.R., E. Suhartatik, and A.K. Makarim 2010.Productivity and yield components of rice as a function of plant population. In: S. Abdulrachman, H.M. Toha, and A. Gani (Eds.). Rice Technology Innovation to Maintain Self-Sufficiency and Encourage Rice Export.National Seminar Proceeding on Rice Research Results 2009, Book 2.Indonesian Center for Rice Research.p.443-450.

[13] Pujiasmanto, 2013. Strengthen our national food security. Professor of the Faculty of Agriculture, SebelasMaret University (UNS).Surakarta http://www.uns.ac.id. Sauki A, Nugroho. A and Soelistyono.E, 2014.The Effect of Spacing and Inundation on the Sri (System of Rice Intensification) Method on Growth and Yield of Rice (Oryzasativa.L.).Journal of Plant Production, Volume 2, Number 2, March 2014, p.121127.

[14] Supriyanto E., E Syakiroh J., and Wisnu A. 2010.The effect of the legowo planting system and the concentration of liquid supplementary fertilizers on rice growth and production. Biofarm Agricultural Scientific Journal, 13 (8).

[15] Sohel M. A. T., M. A. B. Siddique, M. Asaduzzaman, M. N. Alam, & M.M. Karim, 2009. Varietal Performance of Transplant Safe Rice Under Diff [e] rent Hill Densities. Bangladesh J. Agril. Res. 34 (1): 33 - 39. Accessed July 25, 2011.

[16] Uphoff, N. 2002.Presentation for C on Raising Agricultural Productivity in the Tropics. Biophysical Challenges For Technology And Policy; The System Of Rice Intensification Developed In Madagascar. 8 things.

https://dx.doi.org/10.22161/ijeab.51.26 *ISSN: 2456-1878*

Effect of dietary incremental levels of flaxseed supplementation on productive performance of lactating Damascus goats

Meteab M. I[1], El-Sayed H. M.[2], Abeer.M. EL-Essawy[1], Nassar M.S.[1], El-Bordeny N. E.[2]

[1]Animal and poultry Nutrition Dept., Animal and poultry Divi., Desert Research Center., Mataryia, 11753, Cairo, Egypt.
[2]Animal Production Dept., Fac. of Agric., Ain-Shams Univ., 68 Hadayek Shoubra 11241, Cairo, Egypt.

***Abstract*— *It is believable that supplemental essential fatty acids can change the fatty Acid (FA) composition in the milk. Feeding flaxseed to dairy animals improves milk production and milk quality, resulting in healthier milk for consumer. So, the objective of our study was to evaluate the effect of inclusion of ascending levels of flaxseed in Damascus goat's ration on performance and milk composition. Twenty-four of lactating Damascus goats (39.60 ±0.50 kg weight and 2-3 years old) were divided into three groups (randomly, eight animals each). The basic diet of control group (T1) consisted of 56.67% concentrate feed mixture (CFM) and 33.33% alfalfa hay and supplemented with 10% full fat soya, while the other groups were supplemented with 5% flaxseed + 5% full fat soya (T2) and 10% flaxseed (T3), respectively. Inclusion of higher level of flaxseed (10%) in goat's ration increases dry matter intake (DMI) with the positive effect on digestibility of most nutrients. In addition, rumen fermentation was affected with increased fat supply where levels of total volatile fatty acids (TVFA's) and ammonia-N (NH_3-N) are increased with reduced rumen pH values in animals fed on T3. In this study, significant increase of blood plasma total protein, globulin, albumin, urea and high-density lipoprotein concentration, whereas significant decrease of triglycerides, cholesterol and Low-density lipoprotein concentration in response to higher supplemental fat than T1 and T2. Goats supplemented with higher level of flaxseed recorded higher body weight, milk yield and fat corrected milk (FCM) yield, milk fat, protein and total solid content than the other groups (T1 and T2). In conclusion, higher flaxseed supply in dairy Damascus goat's diets resulted in improved total tract digestibility, feed efficiency and rumen fermentation parameters and milk production, milk composition while reduced blood lipids.*

Keywords*— *Flaxseed, Damascus goat, digestibility, feed intake, milk production.

I. INTRODUCTION

Recently there has been an interest in using flaxseed in animal rations as it can be used to alter the fatty acid composition of milk products and improve animal performance, therefore, provide functional health benefits for the consumer. Flaxseed is an excellent source of high-quality protein and energy for ruminants **(Neveu *et al.*, 2014)**. The oil content in flaxseed ranges between 40% and 45% **(Mohamed, 2013)**. Flaxseed is a great source of essential fatty acids, which contains approximately 50%–70% of α- linolenic acid (ω-3 fatty acids) **(Xu *et al.*, 2013)**. So, there has been an increasing interest in use of oilseeds to improve the ruminant dairy products, because of the increasing consumer awareness of food healthiness. Although, **Benchaar *et al.* (2012)** reported that the inclusion of supplemental fat did not decrease or increase the nutrient digestibility, but **Piantoni *et al.* (2013)** found that palmitic acid enhanced the total tract digestibility of NDF, organic matter and CP. **Nawaze and Ali (2016)** suggested that generally fat inclusion in diet increased the milk production clearly compared with control diet while **Gargouri *et al.* (2006)** demonstrated that up to certain level of fat inclusion in diet leads to increased milk production and after that level of milk yield decrease. On the other hand, increasing inclusion level of fats in the diets of the ewes and goats resulted in linear increase of milk fat content **(Casals *et al.*, 2006)** whereas, decreased the milk protein in cows and ewes but not in goats **(Nawaz and Ali, 2016)**.

https://dx.doi.org/10.22161/ijeab.51.26 *ISSN: 2456-1878*

This study aimed to evaluate the effect of increment of flaxseed supply levels (two levels versus control) in Damascus goat's ration on its productive performance during lactation period.

II. MATERIALS AND METHODS

This experiment was conducted at the Mariout Research Station (30 km to Alexandria) and labs of animal nutrition department, Desert Research Center (DRC) , El-Matarya , Cairo, Egypt.

The experimental animals, design and rations

Twenty-four Damascus goats (39.60 ±0.50 kg and 2-3 years) were randomly divided into three groups (eight animals each). All of the experimental groups were fed on 90% basal diet that consisted of 56.67% concentrate feed mixture (CFM) and 33.33% alfalfa hay) and supplemented with one of these supplements:10% full fat soya (T1), 5% flaxseed + 5% full fat soya (T2) or 10% flaxseed (T3), respectively. Three experimental rations were formulated to cover goats requirements according to **(NRC 1981)**. The chemical composition of the feed ingreadents and the experimental rations are presented in Table (1). Complete rations (concentrate + alfalfa) were offered twice daily at 7 am and 4 pm in quantities sufficient to allow free choice access to the ration, and animals have free access to clean fresh water. The animal weighed biweekly befor morning feeding and the orts were determined.

Table 1. Chemical composition of feed ingredients and the experimental rations (% on DM basis).

	feed ingredients				Complete rations		
Items	Flaxseed	Concentrate	Full fat soya	Hay	T1	T2	T3
Dry matter, %	95.93	90.76	93.58	92.66	91.68	91.79	91.91
organic matter, %	96.04	92.47	92.75	87.15	89.88	90.07	90.26
Ash, %	3.96	7.53	7.25	12.85	10.12	9.93	9.74
Crude protein, %	20.06	16.73	37.64	16.28	20.37	19.38	18.40
Ether extract, %	40.24	3.30	16.28	2.19	4.61	5.91	7.21
Crude fiber, %	28.55	13.16	10.23	30.83	20.46	21.43	22.40
Neutral detergent fiber, %	48.43	30.76	31.89	46.31	39.33	40.18	41.03
Acid detergent fiber, %	32.32	14.71	13.65	30.78	21.77	22.76	23.75
Nitrogen Free Extract, %	7.20	59.30	28.60	37.80	44.44	43.35	42.25
Non fiber carbohydrate, %	--	41.68	6.94	22.37	25.57	24.6	23.62

Oilseeds Fatty acids analysis

Fatty acids contents of soybean and linseed were analyzed according to **AOAC, (2000)** using Ultra Gas Chromatographs (Table 2).

Digestibility trials

A digestibility trial was performed at the end of lactation period and samples were taken through 45 days of lactation period. The feces were collected using fecal grab samples method from all doses, three times daily (7.00, 14.00 and 18.00) for three consecutive days. Acid-insoluble ash was used as an internal marker to estimate fecal output and nutrient digestibility. The digestibility coefficient of a given nutrient was calculated according to the following formula **(Van Kulen and Young, 1977):**

$$Digestibililty = 100 - \frac{\%indicator\ in\ the\ feed}{\%\ indicator\ in\ the\ feces}\ x\ \frac{\%\ nutrient\ in\ the\ feces}{\%\ nutrient\ in\ the\ feed}$$

Rumen liquor samples

Rumen liquor samples were randomly collected from four goats within each group using a stomach tube as described by **Khattab *et al.* (2011)** before the morning feeding (zero time), 3 and 6 h after the morning feeding. pH was immediately determined using pH meter (Gallen Kamp pH Stick pH K-120 – B). Then samples were filtered through two layers of sheethcloth, into 25 ml glass bottles with adding few drops of toluene to stop fermentation and 5 ml of paraffin oil just to cover the surface and kept in deep freeze (-18°C) till subsequent analysis.

https://dx.doi.org/10.22161/ijeab.51.26 *ISSN: 2456-1878*

Table.2: Fatty acids content (% of total) of the experimental oilseeds.

Fatty acid	Oilseeds	
	Flaxseed	Full fat soya
C16:0, Palmitic acid	5.52	13.90
C18:0, Stearic acid	4.90	5.72
C18:1n-9, Oleic acid	19.4	23.6
C18:1n-7, Vaccinic acid	0.74	1.30
C18:2n- 6, Linoleic acid	14.73	50.36
C18:3n-4,	0.20	ND
C18:3n-3, Linolenic acid	53.4	4.53
C20:0, Arachidic acid	0.18	0.40
C20:1n-9, Gadolic acid	0.13	ND
C22:0, Behenic acid	0.15	0.19
Non identified fatty acids	0.65%	ND

ND: not detected

Blood samples

At the end of the experimental trial, blood samples were taken from 4 animals for each group (the same animals were used to get rumen liquor content sample). A sample of 10 ml of blood per animal was withdrawn from the jugular vein before morning feeding. The blood samples were directly collected into vacationer tubes (containing EDTA as an anti-coagulant). The blood plasma was obtained by centrifuging the blood samples soon after collection at 4000 rpm for 15 minutes. Blood plasma was transferred into a clean dried glass vials and then stored in deep freezer at -18º C for subsequent specific chemical analysis.

Milk samples

Daily milk yield (DMY) was individually recorded weekly after colostrum period, up to 12th week of lactation. Doses were kept away from their kids for 12 h (9 pm: 9 am) (overnight), and then one teat was hand milked while the second teat was left for suckled kids. The daily milk yield was determined in two consecutive days the first for left teat and the second for right teat. Consequently, DMY was estimated as an average of the two teats. Milk was multiplied by 4: (2 teats X 2 (two half day) to complete 24 h) **(Alsheikh, 2013).** Milk samples were obtained weekly from each goat for 12 weeks and stored in glass bottles (50 ml) then analyzed to determine milk composition.

Analytical methods

Feedstuffs and fecal analysis

Samples (feeds and feces) were oven-dried (55° C for 72 h), then ground in welly mill fitted with a 1 mm screen (local manufacture). Feeds and fecal samples were subjected to proximate chemical analyses crude protein (CP), crude fiber (CF), ether extract (EE) and Ash according to **AOAC (2000)** while nitrogen free extract (NFE) was calculated by difference. Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were determined in sequential procedures of **Van Soest *et al.* (1991)**, analysis using Ankom^{200} apparatus (Ankom Technology Corp., Fairport, NY) filter bag technique. Non-fiber carbohydrate (NFC) was calculated according to the following formula:

NFC (%) = 100− (%NDF+ %CP + %fat + %ash) (**NRC, 2001**).

Determination of basic rumen fermentation parameters

The pH of rumen liquor was immediately recorded using Gallen Kamp pH Stick pH K-120 – B. quantitative analysis of ammonia concentration was carried out by a modified Nessler's method modified by **Szumacher-Strabel *et al.* (2002)** and total volatile fatty acids (TVF's) were determined by steam distillation according to **Warner (1964)**.

Biochemical analysis of blood plasma

Blood serum samples were analyzed using commercial kits (Human Co. Germany). Total protein, albumin, urea, and creatinine were used as indicators for kidney function, while alanine aminotransferase (ALT) and aspartate aminotransferase (AST) were used as indicators for liver function and lipid profile (triglycerides (TG), cholesterol, high-density lipoprotein (HDL)-cholesterol, low-density lipoprotein (LDL)-cholesterol and total lipids) as indicators for fat mobilization. All measurements were done using Jenway spectrophotometer (UK) and the kits purchased from Human Co. Globulin concentration was calculated by

subtraction of total plasma protein and plasma albumin. The albumin /globulin (A/G) ratio was calculated.

Milk analysis

Milk samples were analyzed for total solids, fat, total protein and lactose by infrared spectrophotometry (Foss 120 Milko-Scan, Foss Electric, Hillerød, Denmark). Solids-not-fat (SNF) was calculated by difference. Fat corrected milk (4% fat) was calculated by using the following equation according to Gaines (1928):

FCM = 0.4 milk yield (gm) + 15 fat yield (gm)

Statistical analysis

Data were statistically analyzed using (**SAS, 2006).** Separation among means was carried out according to Duncan Multiple Range test **(Duncan, 1955).** Data of body weight changes, digestibility and blood parameters were statistically analyzed according to the following model: $Y_{ij} = \mu + T_i + e_{ij}$, Where y_{ij} = represents observation, μ=: the overall mean, T_i = effect of treatment (experimental group), e_{ij}: experimental error. While the data of rumen fermentation parameter and milk production were statistically analyzed according to the following model: $Y_{ij}= \mu + T_i +S+ an(t) +S*T + e_{ij}$. Where: Y_{ij}= the observation on the I^{th} treatment, μ = Overall mean, T_i = Effect of the I^{th} treatment, S = Effect of the period, an (t) = Effect of the animal in the treatment and e_{ij}= Random experimental error.

III. RESULTS AND DISCUSSION

Oil seeds fatty acid composition:

Fatty acid (FA) profiles of the two oilseeds are completely different as indicated in Table (2). it is evident that linseed is the richest source of linolenic acid (C18:3n-3) (53.4% of the total fatty acids) followed by oleic (C18:1n-9), linoleic (C18:2n-6), palmitic (C16:0), then stearic acid (C18:0) as (19.4, 14.73, 5.52 and 4.90%, respectively). However, soybean is the richest source of linoleic acid (50.36%) and the rest of FA which are oleic, palmitic, stearic and linolenic acids accounting for formed 23.6, 13.9, 5.72 and 4.53% of the total FA, respectively.

Effect of experimental rations on digestibility and nutritive value

Animals supplemented with the highest level of flaxseed (10%, T3) recorded significant higher digestibility of all nutrients (DM, P=0.019, OM, P=.044, EE, P=0.007, CF, P=0.02 except CP showed non significant differences P=0.45 and nitrogen free extract (P=0.056) compared to 0 and 5% levels (T1 and T2) (table 3). improved digestibility with 10% flaxseed supply may be due to that flaxseeds are small, flat and oval-shaped (2×5 mm), therefore, flaxseed may result in higher possibility of escaping from mastication so, increased passage rate from the rumen and packaging the fat and protein in such a way not to negatively affect rumen function, while promoting feed intake, increase the energy content of the diet and gives a partial protection versus microbial attack or reduces the impact of oil on ruminal microbial or both, leading to negligible effect on the digestion of fibers as well as Improved CP digestibility **(Khorasani *et al*., 1992, Syed *et al*., 2012 and Kim *et al*., 2004)**. In this connection, **Dayani *et al*. (2011)** recorded that feeding flaxseed to ruminants affecting rumen function positively and increase the nutrients availability in the small intestine. Also, **Gonthier *et al*. (2004)** reported an increment of total digestibility of organic matter and fiber with extruded flaxseed supply. In addition, flaxseeds are sources of unsaturated fatty acids which, generally, highly digestible compared to saturated fatty acids **(Palmquist and Mattos, 2006)**. Conversely, **Machmüller *et al*. (2000)** did not find any variations in digestibility when they feed lambs 6.7% flaxseeds, **Wachira *et al*. (2000)** with sheep fed 10.5% flaxseeds and **Paula *et al*. (2014)** with oilseeds in Saanen goat diets.

The reduction in CF digestibility in the animals fed ration of T1and T2 (supplemented with 10 and 5% full fat soybean) compared to the animals fed ration of T3 (supplemented with 10 % flaxseed) may be due to that the fats in full fat soy is not protected and affect negatively on rumen function and cellulolytic bacteria which led to decrease fiber digestion on the contrary for flaxseed the fat is protected as indicated by **Khorasani *et al*. (1992); Syed *et al*. (2012) and Kim *et al*. (2004)**

Improved feed digestibility in the present study resulted in significant enhancement of nutritive value as total digestible nutrients (TDN, P= 0.001 % with 10% flaxseed supplementation (T3). However, digestible crude protein (DCP%) increased (P=0.009) with 10% soybean supplementation. This may be due to that the ration containing 10 % soybean recorded higher CP contents (20.37) compared to the other experimental treatments Table (1)

https://dx.doi.org/10.22161/ijeab.51.26 *ISSN: 2456-1878*

Table.3: Effect of feeding experimental rations on digestibility of nutrients during the lactation period.

Items	T1	T2	T3	SE	P
Dry matter, %	56.60[b]	56.12[b]	61.20[a]	1.1071	0.019
Organic matter, %	59.18[b]	60.55[b]	63.29[a]	0.9850	0.044
Crude protein, %	79.05	77.9	81.74	0.9545	0.45
Ether extract, %	62.43[b]	73.02[b]	82.42[a]	3.2858	0.007
Crude fiber, %	38.02[b]	41.36[b]	49.64[a]	2.4066	0.02
Neutral detergent fiber, %	31.92[b]	38.76[b]	43.68[a]	2.5424	0.03
Acid detergent fiber, %	32.28[b]	33.96[b]	41.42[a]	2.1368	0.03
Nitrogen free extract, %	60.84	62.12	62.83	1.2821	0.56
Nutritive value					
Digestible crude protein, %	15.85[a]	14.93[b]	14.93[b]	0.1851	0.0094
Total digestible nutrients, %	56.17[c]	59.55[b]	64.38[a]	1.0220	0.0010

[a] and [b], means with different superscripts in the same row are significant different.

Effect of experimental rations on Feed intake

Results of dry matter intake (DMI) Table (4) showed that supplementation with higher level of flaxseed (T3) resulted in numerically higher dry matter intake (DMI) during the lactation period. This may be attributed to the increment in nutrient digestibility (table 4) which promote rumen discharge consequently force the animal to eat a lot.

Table.4: Effect of feeding experimental rations on feed intake during the lactation period.

Items (g/h/d)	T1	T2	T3
Dry matter intake	2210.62	2213.94	2284.81
Total Digestible Nutrients intake	1241.73	1318.46	1470.91
Crude protein intake	443.18	424.25	417.49
Digestible Crude protein intake	350.34	330.49	341.23

In this connection**, Drouillard *et al.* (2002)** recorded that inclusion of 10% flaxseed in lactating cattle diets led to increase feed intake. However, **Silva-kazama *et al.* (2010)** reported a reduction in dry matter intake when feeding lactating goats on oilseeds and **Benson *et al.* (2001)** attributed this reduction to duodenal availability of fatty acids can decrease feed intake**.**

The goats fed on 10% flaxseeds recorded numerically higher TDN intake compared with the other experimental groups (T1 and T2) in response to increase TDN content for ration of T3 compared to T1 and T2 (table 4). On the other hand, the animals of T1 recorded higher crude protein intake (CPI) and digestible crude protein (DCPI) compared to T2 and T3 as a result of increased CP content for ration T1 (table 1) as well as increase of DCP content for T1 compared to T2 and T3 (table 4).

Effect of experimental rations on rumen fermentation parameters

Concerning ruminal fermentation parameters Table (5) it is clear that 10% supply of flaxseed improved rumen fermentation where total volatile fatty acids (TVFA's) and ammonia concentration increased as a mean value due to the effect of treatment compared to the other groups (T1and T2). These results disagree with the results of **Broudiscou et al. (1994)**, who reported a decrease in total VFA concentration in sheep supplemented with 6% of flaxseed oil in a forage-based diet. Also, in this connection **Ueda et al. (2003)** observed higher ruminal ammonia with flaxseed

oil supply to dairy cows, whereas **Doreau et al. (2009)** reported no change in ammonia concentration with flaxseed oil supply in dairy cows. Contradicting with these results, **Ikwuegbu and Sutton, (1982)** and **Broudiscou et al. (1994)** reported a decrement in ammonia concentration in sheep supplemented with different levels of flaxseed oil.

This controversial in results may be due to the level of supplement and it's form (oil or seed), experimental animal, experimental ration or experimental conditions as whole. Regarding the ruminal pH level, it is decreased significantly with higher level of flaxseed supply and this may be related to increased production of TVF'S resulting in decrease in pH value.

Table.5: Effect of feeding experimental rations on rumen fermentation parameters during the lactation period.

Items	T1	T2	T3	mean(time)	SE
Total volatile fatty acids meq dl^{-1}					
0h	7.95	5.88	9.70	7.84^{b}	0.2245
3h	5.38	7.90	8.63	7.3^{b}	0.2245
6h	7.23	7.48	12.33	9.01^{a}	0.2245
Mean	6.85^{b}	7.083^{b}	10.22^{a}		
Ammonia concentration, mg dl^{-1}					
0h	5.025	5.3	6.325	5.55	0.1578
3h	4.225	6.4	5.95	5.53	0.1578
6h	4.575	5.65	6.925	5.72	0.1578
Mean	4.61^{b}	5.78^{b}	6.40^{a}		
pH value					
0h	6.95	6.83	6.77	6.85a	0.0818
3h	6.58	6.55	6.25	6.46b	0.0818
6h	6.68	6.43	6.41	6.51b	0.0818
Mean	6.74^{a}	6.61^{a}	6.47^{b}		

[a] and [b], means with different superscripts in the same row are significant different.

Effect of experimental rations on blood parameters

Blood plasma concentrations of total protein (TP), albumin and globulin (Table 6) were increased significantly (P<0.0001) with higher flaxseed level (T3) compared to the other experimental groups (T1 and T2). This may be due to that T3 recorded the highest CP digestibility (table 3) and the highest DMI and TDNI compared to the other experimental groups (table 4). **Kumar *et al.* (1980)** and **Bush, (1991)** postulated that blood plasma total proteins concentration reflects the nutritional status of the animal and reported a positive correlation between blood total proteins concentration and dietary protein level. Moreover, protein fractions of flaxseed composed of albumin, globulin, glutelin and prolamin where the globulin being the major fraction **(Oomah and Mazza, 1993)**.

Blood plasma levels of lipid profile were mainly affected by flaxseed level. Concentrations of triglycerides (TG), cholesterol, total lipids and low-density lipoproteins (LDL) were decreased significantly (P<0.0001) with increasing the level of flaxseed supply compared with zero flaxseed supply. These may be due to the higher (ω-3) fatty acids concentration in flaxseed compared to soybean seed (53.4 Vs 4.53, Table **2**). In this connection **Harris *et al.* (1997)** found that (ω-3) fatty acids reduce plasma triglyceride levels, by inhibiting the synthesis of low-density lipoprotein and triglycerides in the liver. The present results supported this concept because about 53% of fatty acids content of flaxseeds are α-linolenic acid (ω-3) (table 2) that inhibiting the synthesis of very low-density lipoprotein cholesterol and triglycerides in the liver. Consequently, feeding whole flaxseed increased blood concentrations of (ω-3) fatty acids and decreased the ω-6 fatty acid level in blood **(Petit, 2002)**. It is also possible to attribute the reduction of cholesterol and triglycerides levels to flaxseed CP content, where **Bhathena *et al.* (2002)** found that flaxseed proteins were effective in lowering plasma cholesterol and triacylglycerol levels compared to soybean and casein proteins in obese rats. The gradual increase

(P=0.0001) in level of high-density lipoprotein (HDL) in blood plasma of animals fed on rations supplemented with 5 and 10% flaxseed in the current study was matching with the reduction of cholesterol, triglycerides and LDL levels because HDL removes fats and cholesterol from cells including within artery wall and transport it back to the liver for excretion or reutilization **(Peter, 2005).**

Table.6: Effect of feeding experimental rations on some blood plasma parameters during lactation period.

Items	T1	T2	T3	SE	P value	Normal rang
Total protein, g/dl	7.12^{c}	7.40^{b}	8.52^{a}	0.23	<.0001	6.4 -7.8
Albumin, g/dl	4.66^{b}	4.59^{b}	5.14^{a}	0.18	<.0001	2.4 -4.4
Globulin, g/dl	2.45^{c}	2.80^{b}	3.38^{a}	0.25	0.0022	Ne
Urea, mg/dl	39.13^{b}	41.21^{b}	51.17^{a}	3.28	<.0001	15-50
Creatinine, mg/dl	0.84^{a}	0.65^{b}	0.54^{c}	0.07	<.0001	0.9 -1.8
Total lipids, mg/dl	868.0^{a}	866.4^{a}	745.6^{b}	27.6	<.0001	Ne
Cholesterol, mg/dl	210.8^{a}	188.6^{b}	174.9^{c}	5.3	<.0001	150-225
Triglycerides, mg/dl	111.2^{a}	99.6^{b}	90.5^{c}	4.34	<.0001	40-140
High-density lipoprotein, mg/dl	59.7^{c}	66.1^{b}	73.9^{a}	2.74	0.0001	Ne
Low-density lipoprotein, mg/dl	176.1^{a}	159.2^{b}	141.4^{c}	5.18	0.0001	Ne
Aspartate aminotransferase, Units / ml	33.97	34.25	36.47	2.47	0.2281	Up to 40
Alanine aminotransferase, Units / ml	14.94^{b}	15.28^{a}	15.55^{a}	0.15	0.0003	15- 52

a and b mean with different superscripts in the same row are significant different. Normal rang: http://goat-link.com/content/view/204/194/#.XFgUrlwzbIU, ne: not estimated.

Blood urea concentration was increased in animals fed on ration supplemented with 10 % flaxseed compared to the animals fed ration supplemented with zero and 5% flaxseed (Table 6). This increase in urea concentration was supported by the increased CP digestibility (Table 3) as an indicator to improved protein metabolism and improved N utilization with increasing flaxseed level. These results are also supported with higher levels of plasma total protein as an indicator for improved protein metabolism in liver. In this line, **Sharma *e t al.* (1972)** reported lower urea N concentration is usually reported with decreased N digestibility and vice versa.

Regarding creatinine levels, animals fed on T1 recorded significantly higher levels of creatinine than other treatments (T2 and T3), but all values were within the normal range indicating normal renal function. Blood plasma level of aspartate amino transferase (AST) was similar among treatments while flaxseed supply stimulates (P<0.01) blood alanine amino transferase (ALT) activity and its highest level was recorded in goats supplemented with higher level of flaxseed (T3) compared with other treatments although ALT activity lies within the normal range in all treatments. **Nudda *et al*., (2013)** agree with the present findings and they found that inclusion of extruded linseed in dairy goat's diets did not affect renal and hepatic function biomarkers in serum except AST and ALT which tended to differ.

Milk yield, Composition and feed conversion ratio

Data of Table, (8) showed the effect of experimental treatment on milk yield and its composition. Introducing higher level of flaxseed in goats diets (T3) increased milk yield (P<0.01) and improved its composition compared with milk of goats fed T1 or T2 rations. This may be due to increased DM and TDN intake (table 5), improved nutrients digestibility (DM, OM, CP, CF, NDF and ADF) in goats fed on T3 ration (Table 4), leading to increased nutrients availability for milk constitutes synthesis. Similar observations were reported by **Chilliard and Ferlay, (2004)** who generally observed that increase dietary lipids led to increase milk yield, **Gomez-Cortes *et al*. (2009)** in ewes**, Hurtaud *et al*. (2010)** in dairy cows and **Kholif *et al*. (2011)** in dairy buffaloes with flaxseed. Also, higher fat corrected milk (FCM) and fat content% (P<0.01) in milk of T3 fed goats were definitely attributed to high fat content of

https://dx.doi.org/10.22161/ijeab.51.26 *ISSN: 2456-1878*

flaxseed consequently high energy source. Moreover, **Bernard *et al.* (2009); Bionaz *et al.* (2012)** found that fats as dietary supplements encourage the nutrient toward the mammary gland instead of toward fat deposition in the adipose tissue and activate the lipogenic gene expression at mammary gland, leading to an increase of milk fat secretion. Indeed, according to **Zenou and Miron, (2005) and Schwab *et al.* (2006)** increased fiber digestibility in goats fed on T3 diets (table 3) leading to increased milk fat contents. Moreover, use of whole flaxseed as protected fatty acids inside a seed coat did not disturb rumen function so, increased mammary lipogenesis as a result of increased supply of polyunsaturated fatty acid (table 3). **Gargouri *et al.* (2006) and Nudda *et al.* (2013)** consistent with the current results, in sheep Conversely, **Martin *et al.* (2008)** reported decreased FCM yield and fat content on feeding lactating Holstein cows on extruded flaxseed and flaxseed oil diets and they explained these findings by lower DMI and lower digestibility of fiber due to the high level of oil intake. However, **Petit, (2003)** reported no change in milk yield with feeding of (13.3%) whole flaxseed and also **Petit and Cortes, (2010)** when feeding of (72 and 36 g/kg DM) whole flaxseed. Increased milk protein concentration ($P<0.01$) in T3 fed goats may be due to the increased CP digestibility (table 3) and increase blood total protien and albumin (table 6). **Nudda *et al.* (2013)** agreed with the present results where they reported that flaxseed supplementation to Saanen goats led to increased milk protein concentrations as a result to higher protein availability in the intestine. Milk total solids (TS) were increased ($P<0.01$) with increased level of flaxseed supply (T3) than the other groups (T1 and T2). This may be due to the increasing content of fat, protein and lactose in milk (table 8). These results agree with **Silva-Kazama *et al.* (2007)** with dairy cows.

Also, increased percentage of solids not fat (SNF) in the same pattern ($P<0.05$) may be due to increased protein and lactos in milk hence SNF are residual substances after extraction of fat from milk. lactose % results didn't affected in the current study similar to **Miroslava *et al.* (2013)** with goats fed flaxseed. The previous results of the current study indicating general and mostly significant improvement in animal performance (increased DMI, nutrients digestibility, milk yield and all milk macro compounds) with increased supply of flaxseed (T3) so, this improvement associated with the best ($P<0.01$) feed conversion ratio (FCR) either related to milk yield or to fat corrected milk.

IV. CONCLUSION

In conclusion, flaxseed supplementations in Damascus goat's diets during lactation period, lead to improve total tract digestibility, reduced blood plasma lipids and rumen fermentation. Also, flaxseed increase milk production (milk yield and fat corrected milk yield), milk fat content and protein concentration in milk. Finally, flaxseed inclusion (10%) has beneficial impacts on the fat profile of milk producing healthier dairy products for consumers. Further studies should be conducted to obtain the best inclusion level of flaxseed to get more benefits.

Table.8: Effect of feeding experimental rations on milk production, composition and feed conversion ratio.

Item	T1	T2	T3	SE	P
			Milk production		
Milk yield, g/h/d	1335.7^{b}	1300.9^{b}	1542.5^{a}	136.8757	0.0004
Fat corrected milk, g/h/d	572.1^{b}	709.7^{b}	1485.9^{a}	120.342	<.0001
			Milk composition		
Fat, %	2.52^{b}	2.41^{b}	3.74^{a}	0.163	<.0001
Protein, %	2.67^{b}	2.09^{c}	3.33^{a}	0.1893	<.0002
Total Solids, %	11.68^{c}	9.64^{b}	12.08^{a}	2.2374	0.0002
Solids Not Fat, %	7.28^{b}	7.22^{b}	8.34^{a}	0.3093	0.0292
Lactose, %	4.46	4.21	4.58	0.1842	0.2112
			Feed conversion ratio calculation by		
Milk yield, kg/kg DM	2.088^{a}	2.4198^{a}	1.632^{b}	0.4514	0.0028
Fat corrected milk, kg/kg DM	4.22^{a}	4.463^{a}	1.70^{b}	0.6714	<.0001

^{a} and ^{b} mean with different superscripts in the same row are significant different. Feed Conversion Ratio (FCR) calculation based on DMI.

REFERENCES

[1] **Alsheikh, S., (2013).** Influence of age and live body weight on daily milk yield of Zaraibi and Shami goats in Sinai, Egypt .J. Annals of Agricultural Science.,58 (1):1-3.

[2] **AOAC.**, (**2000).** Official Methods of Analysis, th 17 edition. AOAC, Arlington, VA, USA.

[3] **Benchaar, C., Romero-Pérez, G.A., Chouinard, P.Y., Hassanat, F., Eugene, M., Petit, H.V. and Côrtes, C., (2012).** Supplementation of increasing amounts of linseed oil to dairy cows fed total mixed rations: Effects on digestion, ruminal fermentation characteristics ،protozoal populations, and milk fatty acid composition. J Dairy Sci., 95: 4578–4590.

[4] **Benson, J.A., Reynolds, C.K., Humphries, D.J., Rutter,S.M. and Beever,D.E., (2001)** Effects of abomasal infusion of long-chain fatty acids on intake, feeding behavior and milk production in dairy cows. J. Dairy Sci., 84:1182-1191.

[5] **Bernard, L., Bonnet, M., Leroux, C., Shingfield, K.J. and Chilliard, Y., (2009).** Effect of sunflower-seed oil and linseed oil on tissue lipid metabolism, gene expression, and milk fatty acid secretion in Alpine goats fed maize silage-based diets. J. Dairy Sci.,92: 6083–6094.

[6] **Bhathena, S.J., Ali, A.A., Mohamed, A.I., Hansen, C.T. and Velasquez, M.T., (2002).** Differential effects of dietary flaxseed protein and soy protein on plasma triglyceride and uric acid levels in animal models. J. Nutritional Bio.,13(11): 684-689.

[7] **Bionaz, M., Thering, B.J. and Loor, J.J., (2012).** Fine metabolic regulation in ruminants via nutrient-gene interactions: saturated long-chain fatty acids increase expression of genes involved in lipid metabolism and immune response partly through PPAR-a activation. Br. J Nutr., 107:179–191.

[8] **Broudiscou, L., Pochet, S. and Poncet, C., (1994).** Effect of linseed oil supplementation on feed degradation and microbial synthesis in the rumen of ciliate-free and refaunated sheep. Anim Feed Sci Technol., 49: 189–202.

[9] Bush, B.M., 1991. Interpretation of Laboratory Results for Small Animal Clinicians. Oxford Blackwell Scientific Publications, London. pp.515, ref.75.

[10] **Casals, R.G., Caja, G., Pol, M.V., Such, X., Albanell, E., Gargouri, A., Casellas, J., (2006).** Response of lactating dairy ewes to various levels of dietary calcium soaps of fatty acids. J. Anim Feed Sci Technol.,131:312-332.

[11] **Chilliard, Y. and Ferlay, A. (2004).** Dietary lipids and forages interactions on cow and goat milk fatty acid composition and sensory properties. Reprod. Nutr. Dev., 44: 467–492.

[12] **Dayani, O., Dadvar, P. and Afsharmanesh, M., (2011).** Effect of dietary whole cottonseed and crude protein level on blood parameters and performance of fattening lambs. Small Ruminant Research., 97: 48-54.

[13] **Doreau, M., Aurousseau, E. and Martin, C. (2009).** Effects of linseed lipids fed as rolled seeds, extruded seeds or oil on organic matter and crude protein digestion in cows. Anim. Feed Sci. Technol., 150:187–196.

[14] **Drouillard, J. S., Good, E. J., Gordon, C. M., Kessen, T. J., Sulpizio, M. J., Montgomery, S. P. and Sindt, J. J. (2002).** Flaxseed and flaxseed products for cattle: Effects on health, growth performance, carcass quality and sensory attributes. Proc.59th Flax Institute, Fargo, N.D., 72-87.

[15] **Duncan, D.B., (1955).** Multiple range and multiple F tests, Biometrics, 11:(١) 1-42.

[16] **Gargouri, A., Caja, G., Casals, R. and Mezghani, I. (2006).** Lactational evaluation of effects of calcium soap of fatty acids on dairy ewes. J. Small Ruminant Res., 66:1-10.

[17] **Gomez-Cortes, P., Bach, A., Luna, P., Juarez, M. and de la Fuente, M.A. (2009).** Effects of extruded linseed supplementation on n-3 fatty acids and conjugated linoleic acid in milk and cheese from ewes. J. Dairy Sci., 92: 4122–4134.

[18] **Gonthier, C., Mustafa, A.F., Berthiaume, R., Petit, H.V., Martineau, R. and Ouellet, D.R., (2004).** Effects of Feeding Micronized and Extruded Flaxseed on Ruminal Fermentation and Nutrient Utilization by Dairy Cows. J. Dairy Sci., 87, 1854-1863.

[19] **Harris, W.S., Ginsberg, H.N., Arunakul, N., Shachter, N.S., Windsor, S.L. and Adams, M. (1997).** Safety and efficacy of Omacor in severe hypertriglyceridemia. J. Cardiovasc Risk.,4:385-391.

[20] **Hurtaud, C., Faucon, F., Couvreur, S. and Peyraud, J.L. (2010).** Linear relationship between increasing amounts of extruded linseed in dairy cow diet and milk fatty acid composition and butter properties. J. Dairy Sci., 93: 1429–1443.

[21] **Ikwuegbu, O. A. and Sutton, J.D., (1982).** The effect of varying the amount of linseed oil supplementation on rumen metabolism in sheep. Br. J. Nutr., 48:365–375.

[22] **Khattab, H.M., Gado, H.M., Kholif, A.E., Mansour, A.M., and Kholif, A.M. (2011)** The potential of feeding goats sun dried rumen contents with or without bacterial inoculums as replacement for berseem clover and the effects on milk production and animal health. Inter J Dairy Sci., 6: 267-277.

[23] **Kholif, S.M., Morsy, T.A., Abedo, A.A., El-Bordeny, N. and Abdo, M.M., (2011).** Milk production and composition, milk fatty acid profile, nutrients digestibility and blood composition of dairy buffaloes fed crushed flaxseed in early lactation. Egyptian J. Nutr. & Feeds., 14: 385-394.

[24] **Khorasani, G. R., De Boer, G., Robinson, P. H., and Kennelly, J. J., (1992).** Effect of canola fat on ruminal and total tract digestion، plasma hormones, and metabolites in lactating dairy cows. J. Dairy Sci., 75:492–501.

[25] **Kim, C. M, Kim, J. H., Chung, T. Y. and Park, K. K., (2004).** Effects of flaxseed diets on fattening response of

hanwoo cattle: 2. fatty acid composition of serum and adipose tissues. J. Anim. Sci., 17(9): 1246-1254.

[26] Kumar, N., U.B. Singh and D.N. Verma, 1980. Effect of different levels of dietary protein and energy on growth of male buffalo calves. Ind. J. Anim. Sci., 15: 513-517.

[27] **Machmüller, A., Ossowski, D.A. and Kreuzer, M., (2000).** Comparative evaluation of the effects of coconut oil, oilseeds and crystalline fat on methane release, digestion and energy balance in lambs. Anim. Feed Sci. Technol., 85:41–60.

[28] **Martin, J.R., Jouany, J.P., Doreau, M. and Chilliard, Y., (2008).** Methane output and diet digestibility in response to feeding dairy cows crude linseed, extruded linseed, or linseed oil.J.Animal science., 86:2642-2650.

[29] **Miroslava, I., Bohuslav, Č., Luboš,Z., Anna, Š., Kateřina, Š. and Miloslav, Š., (2013).** Effects of flax seed supplementation to lactating goats on milk fatty acid content. Food technology., 2:21-28.

[30] **Mohamed, S. E. E., (2013).** Applying ARIMA to Predict Egypt's Self- Sufficiency Rates of Vegetable Oils During the Period 2011-2010, The 20th Conference of Agricultural Economists, Egypt's Food Security in The Light of International Markets' Risks, The Egyptian Society of Agricultural Economists, October 30-31.

[31] **National Research Council [NRC]., 2001.** Nutrient Requirements of Dairy Cattle. National Academy of Sciences, Washington, DC, USA.

[32] **Nawaz, H. and Ali, M., (2016).** Effect of supplemental fat on dry matter intake, nutrient digestibility, milk yield and milk composition of ruminants. Pak. J Agri. Sci.,53 (1): 271 -275.

[33] **Neveu, C., Baurhoo, B. and Mustafa, k. A., (2014).** Effect of feeding extruded flaxseed with different grains on the performance of dairy cows and milk fatty acid profile. J Dairy Sci., 97:1–9.

[34] **NRC. Nutrient Requirements of Goats., (1981)**. Angora, Dairy‹ and Meat Goats in Temperate and Tropical Countries. National Academy Press, Washington, DC, USA.

[35] **Nudda,G., Battacone, A., Atzori, S., Dimauro, C., Rassu, S. P. G., Nicolussi, P., Bonelli, P., and Pulina, G., (2013).** Effect of extruded linseed supplementation on blood metabolic profile and milk performance of Saanen goats. J. Animal sci.,7(9): 1464–1471.

[36] **Oomah, B. D. and Mazza, G., (1993).** Flaxseed proteins A review. J.Food Chem., 48: 109-114.

[37] **Palmquist, D.L. and Mattos, W.R.S., (2006).** Metabolismo de lipídeos. In: berchielli, t. T. Nutrição de ruminantes. Jaboticabal: funep, 287-310.

[38] **Paula, A. G., Claudete, R.A., Luciano, S.d.L., Maximiliane, A.Z., Francisco, A.F.M., (2014).** Effect of whole oilseeds feeding on performance and nutritive values of diets of young growing saanen goats. Ciênc. Agrotec, 38: 181-187.

[39] **Peter, P., (2005).** "The good cholesterol "; high-Density Lipoprotein. Circulation.,111 (5): 89–91.

[40] **Petit, H. V., (2002).** Digestion, milk production, milk composition, and blood composition of dairy cows fed whole flaxseed. J. Dairy Sci., 85:1482–1490.

[41] **Petit, H. V., (2003).** Digestion, milk production, milk composition and blood composition of dairy cows fed formaldehyde treated flaxseed or sunflower seed. J. Dairy Sci., 86:2637-2646.

[42] **Petit, H.V. and Cortes, C., (2010).** Milk production and composition, milk fatty acid profile, and blood composition of dairy cows fed whole or ground flaxseed in the first half of lactation. Anim. Feed Sci. Technol., 158: 36- 43.

[43] **Piantoni, P., Lock, A.L. and Allen, M.S., (2013).** Palimitic acid increased yields of milk and milk fat and nutrient digestibility across production level of lactating cows. J. Dairy Sci., 96:7143-7154.

[44] **SAS Institute., (2006).** SAS User's Guide: Statistics. Ver 9.0. SAS Institute, Cary, N.C., USA.

[45] **Schwab, E.C., Schwab, C.G., Shaver, R.D., Girard, C.L., Putnam, D.E. and Whitehouse, N.L., (2006).** Dietary forage and nonfiber carbohydrate contents influence B-vitamin intake, duodenal flow, and apparent ruminal synthesis in lactating dairy cows. Journal of Dairy Science., 89:174–187.

[46] **Sharma, H.R., Ingalls J.R. and Mckirdy, J. A., (1972).** Nutritive value of formaldehyde-treated rapeseed meal for dairy calves. J. Anim. Sci., 52:363 –371.

[47] **Silva-Kazama, D.C., Santos, G.T.D., Branco, A.F., Damasceno, J.C., Kazama, R., Matsushita, M., Horst, J.A., dos Santos, W.B.R. and Petit, H.V., (2007).** Production performance and milk composition of dairy cows fed whole or ground flaxseed with or without monensin. J. Dairy Sci., 90: 2928-2936.

[48] **Silva-Kazama, D.C., Kazama, R., Gagnon, N., Benchaa, C.R., Santos, G.T.D., Zeoula, L.M. and Petit, H.V., (2010).** Milk composition, milk fatty acid profile, digestion, and ruminal fermentation in dairy cows fed whole flaxseed and calcium salts of flaxseed oil. J. Dairy Sci., 93: 3146–3157.

[49] **Syed, A. S., Afshar, M. A., Hasan, F. and Jaber, D., (2012).** Importance essential fatty acids (n-6 and n-3) in animal nutrition: I: Ruminant. J. Annals of Biological Research., 3, 2:1161-1176.

[50] **Szumacher-Strabel,M ., Potkanski, A., Kowalczyk, J., Cieslak, A., Czauderna, M., Gubala, M. and Jedroszkowiak, P., (2002).** The influence of supplemental fat on rumen volatile fatty acid profile, ammonia and pH levels in sheep fed a standard diet. Journal of Animal and Feed Sciences., 11(4): 577-587.

[51] **Ueda, K., Ferlay, A., Chabrot, J., Loor, JJ., Chilliard, Y. and Doreau, M., (2003).** Effect of linseed oil supplementation on ruminal digestion in dairy cows fed diets

with different forage: concentrate ratios. J. Dairy Sci., 86: 3999– 4007.

[52] **Van Keulen, J. and Young, B. A., (1977).** Evaluation of acid insoluble ash as natural marker in ruminant digestibility studies. J. Animal Sci., 44, 2: 282-287.

[53] **Van Soest, P. J., Robertson, J. B. and Lewis, B. A., (1991)**. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysac-charides in relation to animal nutrition. J. Dairy Sci., 74:3583–3597.

[54] **Wachira, A. M., Sinclair, L. A., Wilkinson, R. G., Hallett, K., Enser, M.and Wood, J. D., (2000).** Rumen biohydrogenation of n-3 polyunsaturated fatty acids and their effects on microbial efficiency and nutrient digestibility in sheep. J. Agric. Sci., 135:419–428.

[55] **Warner, A.C.L., (1964).** Production of volatile fatty acids in the rumen: methods of measurement. Nutr. Abstracts and reviews., 34: 339- 352.

[56] **Xu, J., Gao, H., Song, L., Yang, W., Chen, C., Deng, Q., Huang, Q., Yang, J. and Huang, F., (2013).** Flaxseed oil and α-lipoic acid combination ameliorates hepatic oxidative stress and lipid accumulation in comparison to lard. Lipids Health Dis., 1 (12), 58.

[57] **Zenou, A. and Miron, J., (2005).** Milking performance of dairy ewes fed pellets containing soy hulls as starchy grain substitute. Small Ruminant Research., 57: 187–192.

https://dx.doi.org/10.22161/ijeab.51.27 *ISSN: 2456-1878*

Major Land uses on acid Sulfate soils of Hau Giang province, Vietnam

Vo Quang Minh, Pham Thanh Vũ, Le Van Khoa, Thai Thanh Du, Le Quang Tri, Tran Van Dung

Department of Land Resources, College of Environment & Natural Resources, Cantho University, Vietnam

Abstract— *In recent years, rice crop intensification, which has put a lot of pressure and led to a great change in soil resources and its distribution. In Haugiang province of Vietnam, under different land uses, the previous soil map (2008) has changed and out of date. The research aims to update the soil map of the province under different land uses. The collection of 175 soil profile description data, and 51 soil analysis profiles. Soil classification was followed WRB 2006. The results showed that two major soil groups were found, in which four diagnostic horizons (Mollic, Umbric, Plinthic, and Sulfuric); one diagnostic property (Gleyic); and one diagnostic material (Sulfidic) were identified, and 15 soil types were classified. The Gleysols soil group have 14 soil types (hamoGL, hamoGL(hu), monplGL), (moGL(ptip), moGL(ntip), moGL(dtip), (umGL(ptio), huGL(ptio), umpplGL(ntio), umGL(ntio), huGL(ntio), umGL(dtio), (mowsGL(ntip), umwsGL(ntio)) total area of 9,551.32 ha, accounting for 59.34%; while Anthrosols soil group have one soil type (RGah) area of 66,252.91 ha, accounting for 40.66%.*

Soil map of the province was updated according to WRB 2006, which pineapple, sugarcane crops have a high tolerance of acidity and fruit crops are mainly on Anthrosol soil group, where acid sulfate soils, low in soil pH and base saturation, are dominated. While rice cultivation is dominated on most of Gleysols soil groups, including alluvial and acid sulfate soils. The acid sulfate soils of the study area have low pH, high acidity, high Al content, and low base saturation, in which crops need high tolerance of low pH such as pineapple, sugarcane, fruit crops, but most of the crops should grow on a raised bed for easy to leach soil acidity and toxicity.

Keywords— ***Soil classification, WRB Acid sulfate Soils, Land uses.***

I. INTRODUCTION

Intensive farming in the Mekong Delta has been growing rapidly in recent years. Potential land exploitation is taking place very strongly. The intensive cropping process has greatly altered soil properties, especially accelerated soil degradation processes that depleted the nutrient supply of crops [5]. Formerly established soil map of the region has been changed but have not been updated and no longer respond to the practical situation [6]. Especially in Hau Giang province, in particular, many land-use models have been developed that bring high profit for people [7]. However, land-use changes and intensive cultivation does not pay attention to the conservation of soil resources, along with the changes in natural conditions, since soil properties have been changed [6]. Then, the soil properties and types in the region need to be updated according to the changing of land uses for further land use planning and recommendation.

II. MATERIAL AND METHODS

Data collection: Collect all soil data (soil map of Hau Giang province in 2008, and current land use map of Hau Giang Province in 2015, soil data analysis of Hau Giang province from Land Resources Department, College of Natural Resources and Environment, Can Tho University.

Soil Survey: 175 soil profiles were described. Selection of sites for soil augering and soil profile descriptions, soil sampling for analysis based on the guidelines included Handbook for soil survey, classification, mapping and land Evaluation of Thai Bat et al., 2015. [4] and Guidelines for soil profile description, FAO (2006) [3].

Soil sampling: Sampling at 51 sites on the surface (Ap) and surface horizons of the actual depth of the soil horizon, to identify the main diagnostic horizons, properties, and material as described in [1] and [2].

Soil Classification Method: Use FAO's World Reference Based System (WRB) to classify soil based on soil diagnostic horizons, properties and materials and the rules of the system [1], [2].

Soil analysis: Soil samples was analysis for soil chemical a soil physics for major parameters in the lab of Department of Soil science, Cantho University for soil diagnostics, properties and materials identification and classification

https://dx.doi.org/10.22161/ijeab.51 *ISSN: 2456-1878*

Soil mapping: Map of soil was updated from previous soil map based on the soil types classified, and contoured from previous soil map (2008), combined with land use status and field observation results.

GIS: Mapinfo software was used to create the soil map.

III. RESULTS

Based on the results of soil survey (175 sites for soil profile description) in 7 districts (Chau Thanh A, Phung Hiep, Long My, Vi Thuy, Vi Thanh, Nga Bay and Long My Town) and 51 soil profiles for soil profile description and soil sampling for soil analysis. The soil map of Haugiang was updated.

Soil chemical properties of some profiles and major diagnostic horizons, properties and material for soil classification are shown in below tables:

Table 1: Soil properties at some sampling sites

Code	Soil groups	pH_{H2O} 1:2,5	EC 1:2,5, mS/cm	Al me/100 g	CEC meq/100g	Organic matter (\$\%)	K exch, meq/100g	Na exch, meq/100g	Ca exch, meq/100g	Mg exch, meq/100g	% Base
HG 99	*Acidic*	*3.79*	*0.558*	8.35	*13.90*	*3.98*	*0.324*	*0.086*	*3.63*	*2.64*	*48.06*
HG 110	*Acidic*	*3.55*	*0.338*	*8.88*	*17.34*	*4.26*	*0.517*	*0.060*	*1.06*	*3.42*	*29.16*
HG 112	*Acidic*	*3.76*	*0.374*	*9.31*	*14.70*	*4.57*	*0.185*	*0.062*	*1.74*	*2.19*	*28.41*
HG 135	*Acidic*	*4.12*	*1.040*	4.86	*16.24*	*15.34*	*0.837*	*0.922*	*1.80*	*5.42*	*55.29*
HG 153	*Acidic*	*3.11*	*0.725*	9.50	*14.84*	*21.56*	*0.568*	*0.144*	*0.28*	*4.75*	*38.69*
HG 91	Alluvial	4.88	0.199	0.466	15.86	4.90	0.206	0.257	7.67	4.83	81.73
HG 95	Alluvial	4.16	0.199	4.14	17.20	3.07	0.372	0.126	5.16	5.82	66.73
HG 108	Alluvial	3.67	1.700	3.79	15.41	3.84	0.411	0.189	5.83	4.11	68.40

Table 2: Diagnostic horizons, properties and materials of major soil group in the study area

No	Soil groups	Soil code	Diagnostic horizons	Diagnostic properties	Diagnostic material	Area (ha)	(%)
1	Alluvial	hamoGL	Mollic	Gleyic		49,313.83	30.26
2	Alluvial	hamoGL(hu)	Mollic	Gleyic		6,777.06	4.16
3	Alluvial	monplGL	Mollic, Plinthic	Gleyic		1,264.69	0.78
4	*Acid sulfate*	*moGL(ptip)*	*Mollic*	*Gleyic*	*Sulfidic*	*821.66*	*0.50*
5	*Acid sulfate*	*mowsGL(ntip)*	*Mollic*	*Gleyic*	*Sulfidic*	*4,946.98*	*3.04*
6	*Acid sulfate*	*moGL(ntip)*	*Mollic*	*Gleyic*	*Sulfidic*	*4,640.34*	*2.85*
7	*Acid sulfate*	*moGL(dtip)*	*Mollic*	*Gleyic*	*Sulfidic*	*2,241.35*	*1.38*
8	*Acid sulfate*	*umGL(ptio)*	*Umbric, Sulfuric*	*Gleyic*		*251.76*	*0.15*
9	*Acid sulfate*	*huGL(ptio)*	*Sulfuric*	*Gleyic*		*8,556.19*	*5.25*
10	*Acid sulfate*	*umwsGL(ntio)*	*Umbric, Sulfuric*	*Gleyic*		*2,071.05*	*1.27*
11	*Acid sulfate*	*umpplGL(ntio)*	*Umbric, Sulfuric, Plinthic*	*Gleyic*		*152.64*	*0.09*
12	*Acid sulfate*	*umGL(ntio)*	*Umbric, Sulfuric*	*Gleyic*		*9.216.82*	*5.66*
13	*Acid sulfate*	*huGL(ntio)*	*Sulfuric*	*Gleyic*		*6,368,98*	*3.91*
14	*Acid sulfate*	*umGL(dtio)*	*Umbric, Sulfuric*	*Gleyic*		*86,63*	*0.05*
15	*Acid sulfate*	*ATgl*		*Gleyic*		*66,252,91*	*40.66*

https://dx.doi.org/10.22161/ijeab.51 *ISSN: 2456-1878*

Table 3: The extent of Soil types by WRB system 2006

	Symbol	Soil type (WRB 2006)	Area (ha)	%
I	**GL**	**Gleysols**	**96,709.98**	**59.34**
1	hamoGL	Hapli - Mollic - Gleysols	49,313.83	30.26
2	hamoGL(hu)	Hapli - Humi - Mollic - Gleysols	6,777.06	4.16
3	monplGL	Molli - EndoPlinthic - Gleysols	1,264.69	0.78
4	moGL(ptip)	Molli - EpiProto Thionic - Gleysols	821.66	0.50
5	mowsGL(ntip)	Molli - HypoSali - EndoProto Thionic - Gleysols	4,946.98	3.04
6	moGL(ntip)	Molli - EndoProto Thionic - Gleysols	4,640.34	2.85
7	moGL(dtip)	Molli - BathiProto Thionic - Gleysols	2,241.35	1.38
8	umGL(ptio)	Umbri - EndoOrthi Thionic - Gleysols	251.76	0.15
9	huGL(ptio)	Humi - EpiOrthi Thionic - Gleysols	8,556.19	5.25
10	umwsGL(ntio)	Umbri - HypoSali - EndoOrthi Thionic - Gleysols	2,071.05	1.27
11	umpplGL(ntio)	Umbri - EpiPlinthi - EndoOrthi Thionic - Gleysols	152.64	0.09
12	umGL(ntio)	Umbri - EndoOrthi Thionic - Gleysols	9,216.82	5.66
13	huGL(ntio)	Humi - EndoOrthi Thionic - Gleysols	6,368.98	3.91
14	umGL(dtio)	Umbri - BathiOrthi Thionic - Gleysols	86.63	0.05
II	**AT**	**Anthrosols**	**66,252.91**	**40.66**
1	ATgl	Gleyic - Anthrosols	66,252.91	40.66

According to table 1 and 2, most of the soils in Haugiang province have low pH, due to acid sulfate with thionic horizon, the occurrence of Jarosite mottles, with high Fe and Al. Soils in Haugiang have low ECe, meaning that soils are low salinity. Based saturation at some profile is less than 50% because of low in base cation.

3.1. Diagnosis horizon, diagnostic properties and diagnostic materials

3.1.1. Diagnosis horizons

Based on the results of the survey, the description and soil analysis of the area showed that there are 4 major diagnostic horizons, according to WRB (2006) definition:

- *Mollic horizon:* The thickness of the soil horizon at survey sites ranged from 20 to 60 cm, dark colour (Chroma ≤ 3), with the base saturation of 55.29 - 81.73% and the organic matter (3.48 - 8.31%). (*Fig 1*)
- *Umbric horizon:* The thickness of the soil horizon at survey sites ranged from 20 to 60 cm, dark colour (Chroma ≤ 3), base saturation ranged from 28.41 to 48.06% organic matter (3.53 - 8.24%). (*Fig 2*)
- *Plinthic horizon*: The survey results show that the Plinthic horizon in Hau Giang province has a depth of 35-80 cm and ends at 70-150 cm depth (*Fig 3*)
- *Thionic horizon*: the results of the soil survey showing that the actual acid sulfate soil (the Munsell colour of mottle is from 2.5Y 8/6-8/8 occurred at a depth of 40-150 cm and end at 100-200cm. Besides, sulfidic soil material is also identified. (*Fig 4*)

Fig 1: Mollic horizon

Fig 2: Umbric horizon

Fig 3: Plinthic horizon

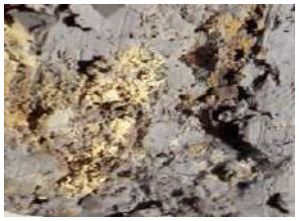


Fig 4: Thionic horizon

3.1.2. Diagnostic properties

According to the classification system WRB 2006, there is one Gleyic diagnostic property identified and used for classifying major soil groups in Hau Giang province. The Gleyic property often occurs at a depth of 40-150 cm. *(Fig .5)*

https://dx.doi.org/10.22161/ijeab.51 *ISSN: 2456-1878*

3.1.3. Diagnostic materials

According to field testing and soil analysis, only sulfidic material identified and often occurs at a depth from 30 to 150cm. *(Fig 6*)

Fig 5: Gleyic property

Fig 6: Sulfidic material

3.2. Major soil groups and soil types in the province

As the above-identified diagnostic horizon, properties, and material, Hau Giang Province has two major soil groups including Gleysols and Anthrosols, which are shown in Table 3 and Figure 7. There are only two major soil groups as Gleysols and Anthrosol, in which Gleysols soil group occupied the largest area (96,709.98 ha, or 50.34%), while Anthrosols soil group is occupied 66,252.91ha (40.00%(

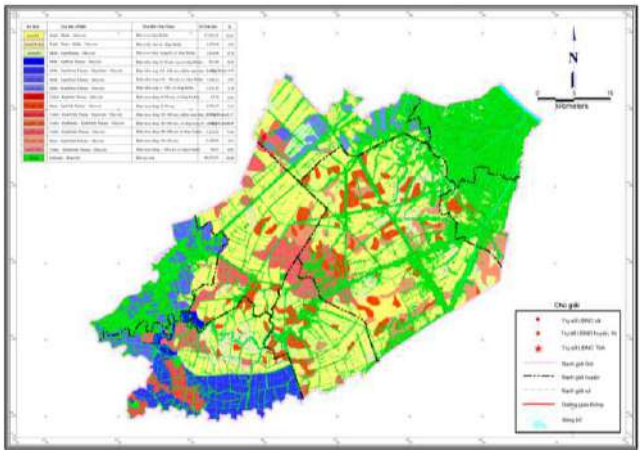


Fig 7: Soil map of Hau Giang province, by WRB system

The Gleysols group in Hau Giang province consists of 14 soil types with a total area of 96,709.98 hectares, accounting for 59.34% of the province's area. In particular, Hapli-Mollic-Gleysols occupied the largest area, 49,313.8ha (30.26%); while Umbri-Bathi-Orthi Thionic-Gleysols occupied the smallest area (86.63ha or 0.05%).

The Anthrosols soil group occupied 66,252.91 hectares, accounting for 40.66% of the province and has only one soil type (Gleyic–Anthrosols).

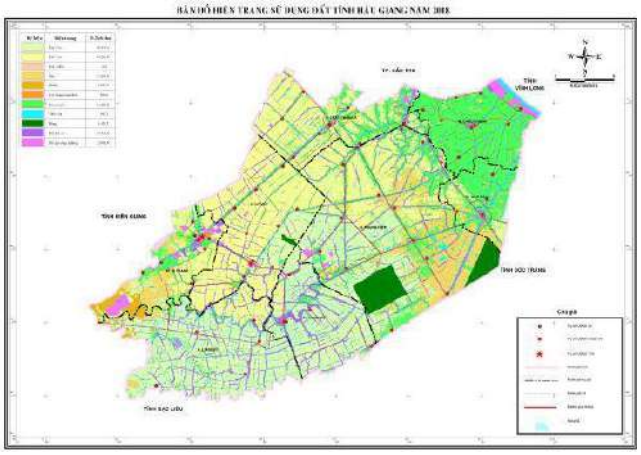


Fig 8: Major land uses map of the study areas

3.3. Major land uses on soils of the study areas

According to Table 4, Fig 7 and 8, most of the soils in the province are alluvial soils, then crops can grow well on these soils, but at some soil types with the occurrence of sulfidic material can release toxicity if oxidized, causing high in toxicity and acidity, low pH, high Al^{3+} and Fe^{2+}, then damage to root crops. These soils have Orthi-Thionic properties, low pH, high toxicity, the result of oxidation of sulfidic material. On these soil types, rice can grow well if under the reduced condition and received freshwater, which can leach toxicity to the canals, and soils get high pH. Otherwise, If freshwater supplied, upland crops can grow well on these soils.

On Anthrosol soil group, Orchard and Upland crops occupied the largest area, (35,240 ha), there is no rice on these soil groups, due to most of the soil is acidic, the occurrent of sulfidic material, low pH, high toxicity. While Pineapple and Sugarcane can tolerate low pH and high toxicity, and soils need to make a raised bed for leaching of toxicity. *(Fig 9*). The rest of the areas are urban, non-Agricultural land, or aquacultural. *(Fig 11)*

On Gleysol soil groups, soils have Gleyic property, it means most of the soils under reduced conditions, higher soil pH and high toxicity, at sometimes of the year, soil can be oxidized to form the soil mottle. Especially, sulfidic soil material is oxidized to release toxicity then the soil has low pH, but in the wet season, because of high rainfall, soil toxicity leached out and rice can grow well on these soils (83,750 ha). However, upland crops such as sugarcane, corn, can growth on these soils if small raised bed created to kept soi dried and toxicity can be drained during the wet season *(Fig 9, 10).*

https://dx.doi.org/10.22161/ijeab.51 *ISSN: 2456-1878*

Fig 9: Sugarcane on acid sulfate soil raised bed

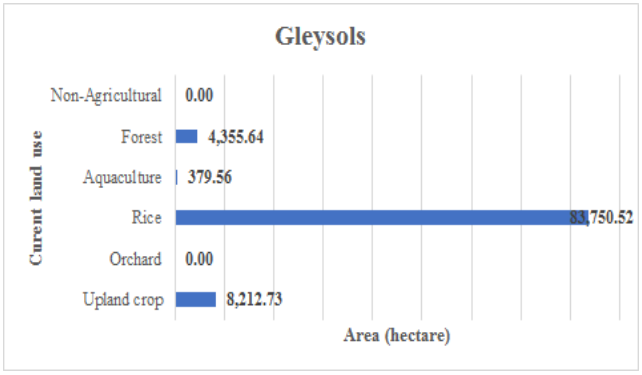


Fig 10: Major land uses on Gleysols soil group

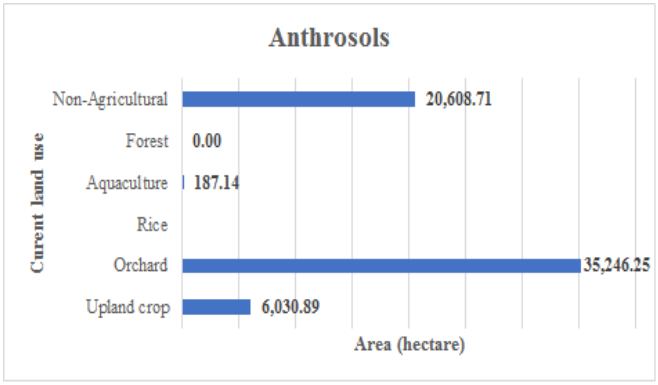


Fig 11: Major land uses on Anthrosols soil group

IV. CONCLUSION

The soil map of the province was updated based on the previous soil map. Two major soil groups identified from WRB system (Gleysols and Anthrosols). Four diagnostic horizons (Mollic, Umbric, Plinthic and Sulfuric), one diagnostic material (Sulfidic) and one diagnostic property (Gleyic) were identified. Within two major soil groups, 15 soil types have been identified: the Gleysols group of 14 soil types (hamoGL, hamoGL (hu), monplGL), moGL (ptip), moGL (ntip), moGL (dtip), umGL huGL, umGL (ntio), huGL (ntio), umGL (dtio), mowsGL (ntip), umwsGL (ntio)) with a total area of 96,709.98 hectares accounting for 59, 34% and Anthrosols with one soil type (ATgl), occupied 66,252.91 ha, accounting for 40.66% or 66%.

Rice cultivation is dominated on Gleysols while Orchard and Upland crops with the raised bed are mainly on Anthrosols soils where acid sulfate soils are dominated.

The acid sulfate soils of the study area have low pH, high acidity, Al toxicity and low base saturation, in which crops need high tolerance of low pH such as pineapple, sugarcane, fruit crops, but most of the crops should grow on a raised bed for easy to leach soil acidity and toxicity.

ACKNOWLEDGEMENTS

This study is funded in part by the Can Tho University Improvement Project VN14-P6, (supported by a Japanese ODA loan), and Ministry of Education support for the annual study, and VLIR (CTU-Belgium) projects, and Department of Science and technology of Hau Giang province.

REFERENCES

[1] FAO, World Reference Center for Soil Resources. 84 World Soil Resource Report, Food and Agriculture Organization of the United Nation Rome, Italy. 1998.

[2] FAO, World Reference Base for Soil Resources 2006. World Soil Resource report. Food and agriculture organization of the United Nation Rome, Italy. 2006a.

[3] FAO. Guidelines for soil profile description (Fourth Edition). ISBN 92-5-105521-1. Rome, Italia. 2006b.

[4] Le Thai Bat, Vu Nang Dung, Bui Thi Ngoc Dung, Do Dinh Dai, Phan Quang Khanh, Do Dinh Sam, Luyen Huu Cu, Phan Van Tu. Handbook for Soil survey, classification, mapping and evaluation. Vietnamese Soil Science Association. Agricultural Publishing house, Hanoi, Vietnam. 2015.

[5] Vo Quang Minh, Nguyen Thanh Vu, Nguyen Thị Ngọc Lan, Phạm Thanh Vu, Thai Thanh Du, Le Quang Tri. Soil fertility evaluation for crop growth in Hau Giang province. Proceeding of Mekong delta soil Science conference. 19 Oct 2018. Cantho University, Vietnam. ISBN: 978-604-954-1069. Cantho University. In Vietnamese. 2018.

[6] Vo Quang Minh, Le Quang Tri, Pham Thanh Vu, Tran Van Dung, Tran Thanh Thang, Thai Thanh Du, The Basis for Conversion of WRB Soil Classification System (1998) to WRB (2006) for Mekong delta soil classification. Vietnamese Soil Science Journal. No 53, 2018. ISSN 2525-2216. In Vietnamese. 2018.

[7] Thai Thanh Du, Ong Van Ninh, Pham Thanh Vu, Tran Van Dung, Vo Quang Minh, Le Quang Tri. Soil classification properties following on WRB 2008 classification system in Hau Giang province. Journal of Soil Science. No 52. 2017. ISSN: 2225 2516. Vietnamese Soil Science Association. 2017. (In Vietnamese)

https://dx.doi.org/10.22161/ijeab.51.28 *ISSN: 2456-1878*

Functions of Exogenously Proline against Negative Effects of Salt Stress in Onion (*Allium cepa* L.)

Dilek Çavuşoğlu[1*], Kürşat Çavuşoğlu[2]

[1*] Department of Plant and Animal Production, Isparta University of Applied Sciences, Isparta, Turkey.
[2]Department of Biology, Süleyman Demirel University, Isparta, Turkey.

Abstract— *The effects of proline on the seed germination, seedling growth (radicle lenght, radicle number and fresh weight), mitotic index, micronucleus frequency and chromosome aberrations of Allium cepa L. germinated under saline conditions were examined in this study. Salt stress markedly inhibited the seed germination and seedling growth of A. cepa L. Moreover, it reduced the mitotic index in the root-meristem cells of the seeds and fairly increased the number of chromosome aberrations and micronucleus frequency which is the simplest indicator, the most effective of cytological damage. On the other, the inhibitive effect of salt stress on the seed germination, fresh weight and mitotic index was significantly decreased with proline application. However, this amino acid was ineffective in reducing of salt damage on the radicle lenght, radicle number, micronucleus frequency and chromosome aberrations.*

Keywords— ***Cytogenetical parameters, onion, proline, physiological parameters, salt stress.***

I. INTRODUCTION

Increasing salinity of agricultural irrigation water together with progressive salinization of agricultural land is of increasing importance to agriculture because it limits the distribution of plants in certain natural habitats and induces a wide range of adverse metabolic responses in higher plants. Salinity stress is one of the most common abiotic factors that inhibit crop growth and productivity by reducing the photosynthetic capacity of plants [1]. High salinity increases the levels of reactive oxygen species (ROS) in plants, such as superoxide radicals, hydrogen peroxide, singlet oxygen and hydroxyl radicals [2]. ROS damage normal metabolism via oxidation of membrane lipids, proteins and plant nucleic acids [3]. Plants develop various defensive mechanisms to cope with salinity-induced damage by compatible solutes as proline and glycinebetaine, and by up-regulating antioxidant enzymes and Na^+/H^+ antiporters [4].

Proline has been known to be involved in the response to a number of environmental stresses such as salt, temperatures, drought, chilling, sorbitol, radiation, heavy metals stress for many years. Proline is an important ozmoregulator that provides of protein integrity and activates of antioxidative enzymes in plants exposed to especially NaCl stress. It is generally accepted that under conditions environmental stresses, proline accumulation serves as a defence against osmotic challenge by acting as a compatible solute. Stress factors, the amount of internal proline in plants cause an increase. Exogenously proline applications are also known to have a positive effect on salt stress tolerance. Plants develop various defensive mechanisms to cope with salinity-induced damage by accumulating such compatible solutes as proline [5, 6]. Proline has been found to protect cell membranes of onion against salt injury [7].

Allium cepa L. (2n=16, chromosomes), the common onion, constitutes a very convenient test system for estimating the harmful effects of chemicals on biological materials. *Allium cepa* test, which is called Levan's test, is one of the most frequently used plant bio-assays. The *Allium* test has been used since it was introduced to evaluate mutagenic effects in the root tips of onions. Additionally, the most important advantage of *A. cepa* bioassay is supported by the very similar of the mutagenic activity of numerous compounds on mammalian cells and *Allium* test cells. This test is now frequently used for laboratory studies [8]. The present study was designed to examine the influences of proline in the reducing of detrimental effects of salt stress on the seed germination, seedling growth, mitotic activity, micronucleus frequency and chromosomal aberrations of *Allium cepa* L.

II. MATERIALS AND METHODS

In this study, *Allium cepa* L. seeds were used. Salt (NaCl) concentration used was 0.125 M. L-proline concentration used in the experiments was 75 mg L^{-1}. L-

proline were obtained from Merck. By a preliminary investigation carried out, firstly it was determined as 0.125 M salt concentration (tried out concentrations of 0.10, 0.125, 0.15, 0.175, 0.20, 0.225, 0.25, 0.275, 0.30 M) which largely preventing the germination of *A. cepa* L. Then it was designated as 75 mg L^{-1} L-proline concentration (tried out concentrations of 1, 5, 15, 25, 35, 45, 55, 65, 75, 100, 200, 300, 400, 500, 600, 700, 800, 900 and 1000 mg L^{-1} dose of L-proline) alleviating the adverse effects of this salt concentration (0.125 M) on the seed germination and seedling growth. The present study has realized in Plant Physiology and Cytogenetic Laboratories of Biology Department in Süleyman Demirel University.

Germination of seeds of *Allium cepa* L. was carried out at a constant temperature (20°C), in the dark in an incubator. Healthy and approximately equal-sized *A. cepa* seeds were selected. Twenty seeds from each treatment group were placed into the plastic containers. The seeds were divided into four groups: Group I (control) was treated with distilled water for 7 consecutive days. Group II was treated with 0.125 M NaCl alone for 7 consecutive days. Group III was treated with a 75 mg L^{-1} dose of proline for 7 consecutive days. Group IV was treated with a 75 mg L^{-1} dose of proline + 0.125 M NaCl for 7 consecutive days. Plastic containers were placed into an incubator for germination. It was assumed that the radicle should be 10 mm long for germination. At the end of the 7th day, after determination of the final germination percentages, radicle numbers were also recorded, and radicle lengths of the seedlings were measured in mm. In addition, the fresh weights in g/seedling were determined. All experiments were repeated 3 times.

After several days, root tips of germinated *A. cepa* were excised (1-1.5 cm segment) for cytogenetic analysis. Then, they were pretreated with saturated para-dichlorobenzene for 4 hrs, fixed in solution of ethanol: acetic acid (3:1) overnight at room temperature and stored at 4ºC in 70% ethanol until used. The root tips were hydrolysed in 1 N HCl at 60°C for 15 min, were stained with Feulgen for 1-1.5 hrs, smashed in a drop of 45% acetic acid and squashed [9]. 24 hrs later, microscopic slides were made permanent by mounting in balsame. The representative of mitotic phases and mitotic aberrations were photographed (500X) with a digital camera (Olympus C-5060) mounted on an Olympus CX41 microscope. Mitotic index, i.e. percentage of dividing cells scored was evaluated by analysing at least 9.000 cells per treatment (approx. 3.000 per slide). Statistical analyses of all parameters were performed by using SPSS program according to DMRT.

III. RESULTS AND DISCUSSIONS

As shown in Table 1, the germination percentage and radicle length of the group III seeds treated with proline statistically showed the same values as the group I (control) seeds germinated in distilled water medium while their radicle numbers and fresh weight partly increased according to ones of the group I seeds. Yan et al. [10] reported that 0.2 mM exogenous proline application increased the fresh weight of two melon cultivars under normal condition. This result is in agreement with the present findings. Deuschle et al. [11] showed that 100 mM exogenous proline application inhibited growth of tobacco cell under normal condition. The discrepany in the findings indicated that the effect of proline may dependent on the differences in treatment times, concentrations used and plant species.

Salt stress showed the restrictive effect on all examined growth parameters. For instance, the group I (control) seeds germinated in distilled water medium displayed germination 100% on the 7th day while this value became 27% in the group II seeds germinated in 0.125 M salinity. In other words, NaCl prevented 73% the germination of *A. cepa* seeds. Salt stress can perform its preventive effect in many ways. It may interfere with seed germination by changing the water status of the seed so that water uptake is inhibited [12]. The present results showing the decrease in the fresh weight and water content of the seedlings in saline medium may be explained by the failure of the roots to receive sufficient water due to the high osmotic pressure of the medium. The inhibitive effect of salt on the radicle length and radicle number may result from reducing cell division, nucleic acid and protein synthesis [13].

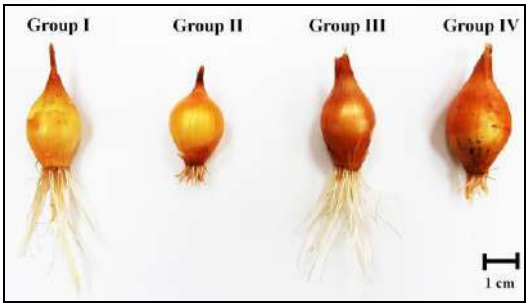


Fig. 1: Root tip cells of Allium cepa showing germination situations at the end of 7 day. Group I (control): distilled water, Group II: 0.175 M NaCl alone, Group III: 75 mg L^{-1} proline and Group IV: 75 mg L^{-1} proline + 0.125 M NaCl. Scale bar = 1 cm

https://dx.doi.org/10.22161/ijeab.51.28 *ISSN: 2456-1878*

Table 1: Effects of proline on some growth parameters of Allium cepa L.

Groups	Growth parameters			
	Germination percentage (%)	Radicle length (mm)	Radicle number	Fresh weight (g/seedling)
Group I	*100 ± 0.0^{c}	58.7 ± 0.7^{b}	45.1 ± 0.7^{b}	10.5 ± 0.3^{b}
Group II	27 ± 2.8^{a}	13.5 ± 1.2^{a}	18.4 ± 1.4^{a}	7.1 ± 0.2^{a}
Group III	100 ± 0.0^{c}	59.3 ± 1.0^{b}	50.1 ± 0.6^{c}	14.0 ± 0.8^{c}
Group IV	70 ± 0.0^{b}	15.0 ± 0.1^{a}	17.6 ± 0.3^{a}	10.1 ± 0.7^{b}

*At the level 0.05 (±SD), the difference between values with the same letter in each column is not significant. Group I (control) treated distilled water, Group II treated 0.125 M NaCl alone, Group III treated 75 mg L^{-1} dose of proline, Group IV treated 75 mg L^{-1} dose of proline+0.125 M NaCl.

Proline application markedly mitigated the inhibitive effect of salt stress on the seed germination. The group IV The fresh weight of the group II seeds grown in 0.125 M salinity was 7.1 g, respectively while this value was 10.1 g in the group IV seedlings treated with proline (Tab. 1). But, the mentioned application was unsuccessful in alleviation of the inhibitive effect of salt stress on the radicle length and radicle number of the seedlings. There are many published studies about the effects of proline on the seed germination and seedling growth under salt conditions until now. However, from these studies, no conclusion could be reached. Thus, proline has been reported to promote [10, 14, 15, 16, 17] or inhibit [11, 18, 19] the germination and growth under salt stress conditions. That proline alleviates salt stress on the seed germination and seedling growth can be understood from the decrease in the salt's osmotic effects. For example, at 0.125 M NaCl medium, proline application partly increased the fresh weights of the seedlings in comparison with the control indicates this probability (Table 1). It reduced the preventive effect of salt on the seed germination and seedling growth by stimulating mitotic activity of the embryo (Table 2). In addition to all these, proline might have been successful in decreasing the inhibitive effect of salt stress on the seed germination and seedling growth by increasing nucleic acid and protein synthesis, by providing stabilization of cell membranes or by raising antioxidant enzyme activities viz. catalase (CAT), peroxidase (POX), superoxide dismutase (SOD) and reactive oxygen species [6, 20, 21].

As a result of our literature studies, although there are a limited number of studies relating to effects of proline on the mitotic index under non-stress and salt stress conditions, no studies were found on the micronucleus frequency and chromosomal aberrations. Therefore, in the present study was carried out to find whether proline is affecting these parameters in normal and saline conditions. The data obtained in this work indicated that seeds treated with proline showed 70% germination (Fig. 1). Proline also continued its success on the fresh weight. the mitotic index in root tip meristems of *A. cepa* germinated in the media containing 0.75 M proline alone increased 96% as compared with group I seeds germinated in distilled water medium. And their micronucleus frequency (11 fold) and chromosomal aberrations (approximately 58 fold) excessively increased according to ones of the group I seeds. In this case, it may be said that some aberrations may result from this imino acid. Mitotic activity expressed as mitotic index decreased at 0.125 M salt concentration (group II) as compared to those of group I (control) samples germinated in distilled water. At the same time, the salt concentration caused an increase on the micronucleus frequency and chromosomal aberrations in root tips of *A. cepa*. For instance, while mitotic index, micronucleus frequency and chromosomal aberrations were 8.2%, 0.0% and 0.0% at control (group I), respectively, they were 0.8%, 9.2% and 12.4% respectively, at 0.125 M NaCl concentration. The inhibitory and cytotoxic effects of salt stress on the mitotic activity are known for a long time [22]. According to some researchers, high salt concentration causes to total inhibition of mitotic activity, micronucleus frequency and chromosomal abnormalities in root-tip cells [23, 24]. On the other hand, proline+NaCl application (Group IV) showed a perfectly good performance in ameliorating the negative effects of salinity on the mitotic index (4.8%). However, the mentioned imino acid application was ineffective in reducing of salt damage on the micronucleus frequency (8.6%) and chromosome aberrations (54.7%). Statistically, all values mentioned here are substantially significant (Table 2).

https://dx.doi.org/10.22161/ijeab.51.28 *ISSN: 2456-1878*

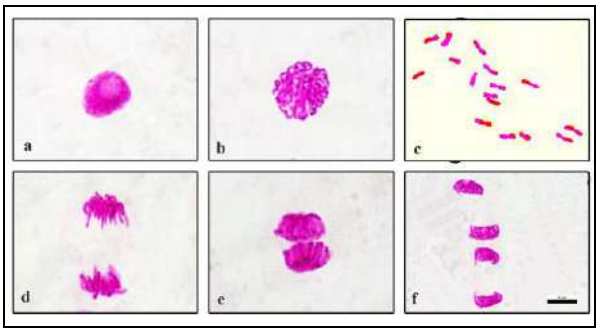


Fig. 2: Normal stages in mitosis a- interphase b- prophase c- metaphase (2n = 16, chromosomes) d- anaphase e- early telophase f- telophases, Scale bar = 10 µm

The normal mitosis phases observed during the microscopic examination of *A. cepa* root tip meristematic cells are shown in Fig. 2 and abnormal mitosis phases are shown in Fig. 3. The chromosomal aberrations noticed in this study were majorly binucleolars in this study. Other chromosomal damages observed in the cells are as follows: micronucleus, irregular prophase, vagrant chromosomes, diagonal at anaphase/telophase, metaphase with chromosome loss, nucleus with nuclear bud, multiple bridge formation in anaphase, adhered chromosomes in metaphase, alignment anaphase, giant cell, chromosomal distrubtions in metaphase, early ball metaphase, ring formation (Fig. 3).

Chromosomal abnormalities (CAs) are changes in chromosomal material or exchange in the structure of the chromosome resulting from breakage. Most of the CAs could affect the fertility, vigour, competitive or yield ability of the exposed plants [25]. Excess proteins and nucleic acids production induced by cytotoxicans result in nuclear buds (Fig. 3b) [26]. As a result of spindle dysfunction, micronucleus (Fig. 3c, o) formation occurs as a result of chromosomal breaks and all chromosomes that do not migrate during the anaphase. Chromosome loss (Fig. 3j) are typically associated with mitotic spindle malfunction. Bridge formation (Fig. 3l) consist of chromatid and/or chromosome breaks, which is indicative the clastogenic effect [27]. İrregular prophase failure (Fig. 3f) might cause chromosome loss when they can not bind to the spindle and therefore are not separated [28]. Ring chromosome (Fig. 3ı) can spontaneously occur after breakage of the chromosomal ends and after the joining of the raw ends of the chromosomes [29]. Diagonal orientation at anaphase / telophase (Fig. 3m, p) was caused by a slight tilt in the spindle apparatus [30].

IV. CONCLUSION

As a result of our literature studies, although there are published studies about the effects of proline application on the seed germination, seedling growth and mitotic index under non-stress and salt stress conditions, current literature data related to the effects of proline application in both normal and saline conditions on the micronucleus frequency and chromosomal aberrations from the cytogenetical studied here have not been encountered. Therefore, this present work was carried out to find whether proline is affecting these parameters in saline conditions or not. As a result, this study showed that proline can significantly increase the activations like the seed germination, fresh weight and mitotic activity under saline conditions. But the mechanisms by which salt inhibits growth are controversial and complex, also they might vary according to cultivar and species. An universal mechanism has still not been established. While the reasons of saltinity have been determined, it is still very poor to understand the mechanisms by which salty prevents plant growth. Therefore, further investigation should be done to learn more about the effect of proline on cell division, cell cycle and germination molecular metabolism. For designing salinity tolerance hypotheses in plants, this literature study can serve to present new conceptual tools.

Table 2: Effect of proline on some cytogenetical parameters of Allium cepa L.

Groups	Mitotic index (%)	Micronucleus frequency (%)	Chromosome aberration (%)
Group I	*8.2 ± 0.3^{c}	0.0 ± 0.0^{a}	0.0 ± 0.0^{a}
Group II	0.8 ± 0.0^{a}	9.2 ± 0.5^{b}	12.4 ± 0.6^{b}
Group III	16.1 ± 0.7^{d}	11.0 ± 1.0^{c}	58.3 ± 1.2^{d}
Group IV	4.8 ± 0.5^{b}	8.6 ± 0.5^{b}	54.7 ± 0.8^{c}

*Shows values with insignificant difference ($P < 0.05$) for each column shown with same letters. Group I (control) treated distilled water, Group II treated 0.125 M NaCl alone, Group III treated 75 mg L^{-1} dose of proline, Group IV treated 75 mg L^{-1} dose of proline+0.125 M NaCl.

https://dx.doi.org/10.22161/ijeab.51.28 *ISSN: 2456-1878*

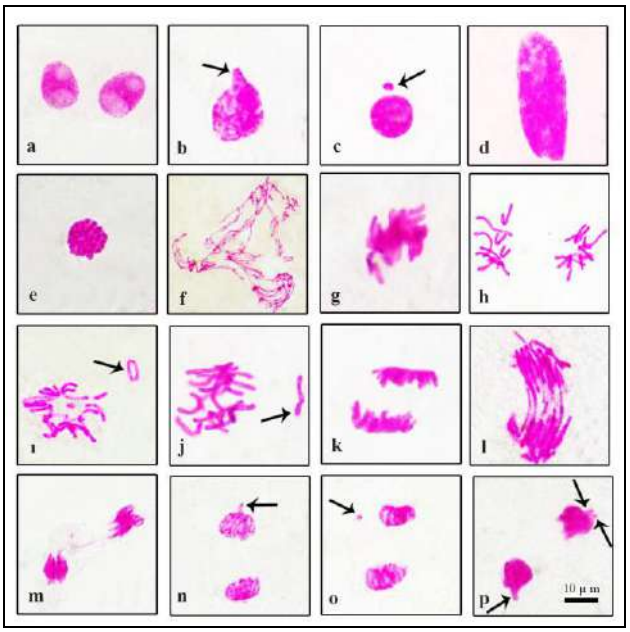


Fig. 3: Main chromosomal aberrations observations in Allium cepa L. meristematic cells, scale bar=10 µm; a: binucleolars b: nucleus with nuclear bud=arrow c: micronucleus=arrow d: giant cell e: early ball metaphase f: irregular prophase g: adhered chromosomes in metaphase h: chromosomal distrubtions in metaphase ı: ring formation=arrow j: metaphase with chromosome loss=arrow k: alignment anaphase l: multiple bridge formation in anaphase m: diagonal at anaphase n: telophase with vagrant chromosome=arrow o: telophase with micronucleus=arrow p: diagonal at telophase with vagrant chromosomes=arrows.

REFERENCES

[1] Subrahmanyam, D. (2008). Effects of chromium toxicity on leaf photosynthetic characteristics and oxidative changes in wheat (*Triticum aestivum* L.). *Photosynthetica*. 46, 339–345.

[2] Larson, R. A. (1995). Plant defense against oxidative stress. *Archives of Insect Biochemistry Physiology*. 29, 175-186.

[3] Hernández, J. A., Ferrer, M. A., Jiménez, A., Barceló, A. R., Sevilla, F. (2001). Antioxidant systems and O_2 /H_2O_2 production in the apoplast of pea leaves. Its relation with salt-induced necrotic lesions in minor veins. *Plant Physiology*. 127, 817-831.

[4] Parida, A. K., Das, A. B. (2005). Salt tolerance and salinity effects on plants: A review. *Ecotoxicol Environmental Safety*. 60(3), 324-349.

[5] Ashraf, M., Foolad, M. R. (2007). Roles of glycine betaine and proline in improving plant abiotic stress resistance. *Environmental and Experimental Botany*. 59, 206–16.

[6] Bayat, R. A., Kuşvuran, Ş., Ellialtıoğlu, Ş., Ustun, A. S. (2014). Tuz Stresi Altındaki Genç Kabak (*Cucurbita pepo* L. ve *C. moschata* Poir.) Bitkilerine Uygulanan Prolin'in, Antioksidatif Enzim Aktiviteleri Üzerine Etkisi. *Turkısh Journal of Agrıcultural and Natural Scıences*. 1(1), 25-33.

[7] Mansour, M. M. F. (1998). Protection of plasma membrane of onion epidermal cells by glycinebetaine and proline against NaCl stress. *Plant Physiology and Biochemistry*. 36 (10), 767–772.

[8] Karaismailoglu, M. C. (2016). Investigation of the potential toxic effects of prometryne herbicide on *Allium cepa* root tip cells with mitotic activity, chromosome aberration, micronucleus frequency, nuclear DNA amount and comet assay. *Caryologia*. 68(4), 323–329.

[9] Sharma, P. C., Gupta, P. K. (1982). Karyotypes in some pulse crops. *Nucleus*. 25, 181-185.

[10] Yan, Z., Guo, S., Shu, S., Sun, J., Tezuka, T. (2011). Effects of proline on photosynthesis, root reactive oxygen species (ROS) metabolism in two melon cultivars (*Cucumis melo* L.) under NaCl stress. *African Journal of Biotechnology*. 10(80), 18381-18390.

[11] Deuschle, K., Funck, D., Forlani, G., Stransky, H., Biehl, A., Leister, D., Graaff, E., Kunze, R., Frommer W. B. (2004). The role of Δ^1-pyrroline-5-carboxylate dehydrogenase in proline degradation. *Plant Cell*. 16, 3413–3425.

[12] Flowers, T. J., Colmer, T. D. (2015). Plant salt tolerance: adaptations in halophytes. *Annals of Botany*. 115(3), 327-331.

[13] Roy, S. J., Negrão, S., Tester, M. (2014). Salt resistant crop plants. *Current Opinion in Biotechnology*. 26, 115-124.

[14] Lone, M. I., Kueh, J. S. H., Jones, R. G. W., Bright, S. W. J. (1987). Influence of proline and glycinebetaine on salt tolerance of cultured barley embryos. *Journal of Experimental Botany*. 38, 479-490.

[15] Gadallah, M. A. A. (1999). Effects of proline and glycinebetaine on *Vicia faba* responses to salt stress. *Biologia Plantarum*. 42, 249-57.

[16] Khedr, A. H. A., Abbas, M. A., Wahid, A. A. A., Quick, W. P., Abogadallah, G. M. (2003). Proline induces the expression of salt-stress-responsive proteins and may improve the adaptation of *Pancratium maritimum* L. to salt-stress. *Journal of Experimental Botany*. 54 (392), 2553-2562.

[17] Hoque, M. A., Banu, M. N. A., Nakamura, Y., Shimoishi, Y., Murata, Y. (2007). Proline and glycinebetaine enhance antioxidant defense and methylglyoxal detoxification systems and reduce NaCl-induced damage in cultured tobacco cells. *Journal of Plant Physiology*. 165, 813-824.

[18] A. B., Garcia, A. J., Engler, M., Van Montagu, A. Caplan, "Effect of NaCl and osmoprotectants on growth inhibition, ion accumulation, and gene expression in rice (*Oryza sativa*)" in Plant Responses to Cellular Dehydration During Environmental Stress, T. J. Close and E. A. Bray, Eds. American Society of Plant Physiologists, 1993, pp 272–273.

[19] Lin, C.C., Kao, C.H. (2001). Cell wall peroxidase against ferulic acid, lignin and NaCl-reduced root growth of rice seedlings. *Journal of Plant Physiology*. 158, 667-671.

[20] Ali, Q., Anwar, F., Ashraf, M., Saari, N., Perveen, R. (2013). Ameliorating effects of exogenously applied proline on seed composition, seed oil quality and oil

antioxidant activity of maize (*Zea mays* L.) under drought stress. *International Journal of Molecular Sciences*. 14(1), 818-835.

[21] Carvalho, K., Freitas de Campo, M. K., Domingues, D. S., Pereira, L. F. P., Vieira, L. G. E. (2013). The accumulation of endogenous proline induces changes in gene expression of several antioxidant enzymes in leaves of transgenic Swingle citrumelo. *Molecular Biology Reports*. 40(4), 3269-3279.

[22] Radic, S., Prolic, M., Pavlica, M., Pevalek-Kozlina, B. (2005). Cytogenetic effects of osmotic stress on the root meristem cells of *Centaurea ragusina* L. *Environmental and Experimental Botany*. 54, 213-218.

[23] Çavuşoğlu, K., Cadıl, S., Çavuşoğlu, D. (2017). Role of potassium nitrate (KNO_3) in alleviation of detrimental effects of salt stress on some physiological and cytogenetical parameters in *Allium cepa* L. *Cytologia*. 82(3), 279-286.

[24] Çavuşoğlu, D. (2019). Physiological and cytogenetical effects of L-Tyrosine in *Allium cepa* L. exposed to NaCl stress. *Fresenius Environmental Bulletin*. 28(12A), 9753-9759.

[25] Kara, M., Tanda, M. A., Ateş, A. (1994). Cytogenetic effects of the insecticide cypermethin on the root meristems of *Allium cepa* L. *Turkish Journal of Biology*. 18, 323–331.

[26] Fenech, M., Kirsch-Volders, M., Natarajan, A. T., Surralles, J., Crott, J. W., Parry, J., Norppa, H., Eastmond, D. A., Tucker, J. D., Thomas, P. (2011). Molecular mechanisms of micronucleus, nucleoplasmic bridge and nuclear bud formation in mammalian and human cells review. *Mutagenesis*. 26, 125–132.

[27] Leme, D. M., Marin-Morales, M. A. (2009). *Allium cepa* test in environmental monitoring: a review on its application. *Mutation Research*. 682, 71–81.

[28] Gisselsson, D., Palsson, E., Yu, C., Mertens, F., Mandahl, N. (2004). Mitotic instability associated with late genomic changes in bone and soft tissue tumours. *Cancer Letters*. 206, 69–76.

[29] Khanna, N., Sharma, S. (2013). *Allium cepa* root chromosomal aberration assay. *Indian Journal of Pharmaceutical and Biological Research*. 1(3), 105-119.

[30] Renjana, P. K., Anjana, S., Thoppil, J. E. (2013). Evaluation of genotoxic effects of baking powder and monosodium glutamate (MSG) using *Allium cepa* assay. *International Journal of Pharmaceutical Sciences*. 5, 132-139.

https://dx.doi.org/10.22161/ijeab.51.29 *ISSN: 2456-1878*

Algal inhibitory efficiency of secondary metabolites of *Tamarindusindica* and *Azadirachtaindica* – A comparative pilot scale study

Mathews P Raj*, Anitha A Abraham, Kasturi Banerjee, Mainak Chakrabarty and Lakshmi Sagar K

Department of Life Sciences, Jain University, Bangalore, India
*Email: pr.mathews@jainuniversity.ac.in

Abstract**—** *This study includes isolation of oil producing algae (Anabaena, Nostoc. Spirulina, Diatom, Volvox, Spirogyra) and subjecting the mixture of algae to the sun dried pulp extract of Tamarindusindica to observe inhibitory effect of secondary metabolites on algal growth. The comparative analysis of inhibiting efficiency was done between the extracts of Tamarindusindica and Azadirachtaindica which proved that the secondary metabolites of Azadirachtaindica are more efficient than the secondary metabolites of Tamarindusindicain inhibiting the growth of algae. 100% and 75% concentration of the crude extracts were used to evaluate the inhibitory effect. Contamination from bacteria and fungi was prevented by maintaining the pH at 8.5. The extract of the sun dried pulp of Tamarindusindica showed inhibitory activity against the above mentioned species of algae.*
Keywords**— Algal inhibition, Tamarindusindica, Azadirachtaindica.**

I. INTRODUCTION

Accelerated eutrophication has been one of the most widespread environmental problems (UNEP). Eutrophication is the enrichment of water by nutrient salts (Nitrate, Phosphate, Potassium) that causes structural changes to the ecosystem such as increased production of algae and aquatic plants, depletion of fish species, general deterioration of water quality and other effects that reduce and preclude use (OECD, 2005). All water bodies are subjected to slow eutrophication but in recent years eutrophication has been accelerated due to anthropogenic activities (Hu *et al*. 2008).

Human development and associated increasing population growth in the watershed area underlie many of the environmental problems occurring in fresh water, transitional (e.g. estuaries and lagoons) and coastal ecosystems (OECD, 2005). Nutrient enrichment (N, P, and K) is one of the most prominent consequences directly related to human activities (Paerl 2006). The nutrient composition has been one of the main factors in excessive proliferation of algae in aquatic ecosystems (Paerl& Huisman 2009). Algal growth promoted by these salts can clog the gills of fish, in addition to anoxic water conditions and death of aquatic life forms (Najem*et al*. 2011). The risk to water quality deterioration is aggravated by the co-dominance of bloom-forming members of the green algae (Chia *et al*. 2016).

Algae like Chlorophytes on their own are not considered being a nuisance, however, nutrient-enriched conditions favor the excessive proliferation of members of this group (Paerl*et al.* 2001). Many types of researches were carried out to control the algal growth by mechanical, physical and chemical methods in addition to bio manipulation (Tesson*et al*. 2014, Zhao *et al*. 2018). All these approaches were unsatisfactory and hence extracts of bioactive compounds from plants that inhibit or prevent algal growth have been in use (Ghorbanian*et al*. 2008). The secondary metabolites of these plants are known to contain antimicrobial properties (Wallace. 2004). Most of the phytochemicals from plant sources such as polyphenols and flavonoids have been reported to have a positive impact on health and cancer prevention (Venugopal*et al.* 2012).

The excessive production of oxygen radicals during algal metabolism is known, especially when they are exposed to

stress conditions (Zhang *et al*. 2013). The presence of bioactive secondary metabolites in plants induce high production of compounds like nitric oxide and H2O2, which have the potential to inhibit antioxidant enzyme activity (Clark *et al*. 2000; Qiao*et al*. 2014). The mechanism of action of these phytochemical extracts may be via lysing the cell, increasing permeability of the cell wall and membrane, inhibition of protein and DNA synthesis and/or by inhibiting the transport of nutrient across the cell wall or membrane (Stewart *et al*. 1979).

Tamarindusindica is a medicinal plant belonging to the family Fabaceae. It has been used as a medicinal plant for centuries; its fruits being the most valuable part. It contains majorly flavonoids, alkaloids and polyphenols (Arranz*et al*. 2010) and has exhibited an inhibitory effect against various organisms (Okoh*et al*. 2017).

Many studies have been done to prevent the growth of algae using the secondary metabolites of various medicinal plants in addition to various physical as well as chemical methods. However, these chemical methods have been causing harmful effects on the environment.

This study explores the potential of *Tamarindusindica* extract to inhibit the proliferation of algae in an environmentally friendly way and compare its efficiency of inhibition with *Azadirachtaindica*leaf extract.

II. MATERIALS AND METHODS

SAMPLE – Algae (lake water), *Tamarindusindica*(fruit), *Azadirachtaindica*(leaves).

GLASSWARE REQUIRED – Petri plates, conical flasks, beakers, glass slides, pipettes.

Instruments / apparatus – Compound microscope, autoclave, centrifuge, colorimeter, Soxhlet apparatus, cork borer, micropipettes.

Chemicals used - Methylene blue, Safranin, 1N NaOH, Ethanol, 0.1 N HCl, acetone, Triple distilled water, conc.HCl, conc. H_2SO_4.

Other Requirements – Forceps, needles, pH paper, centrifuge tubes, plastic trays, sample bottles, spatula, blotting paper, filter paper, cotton, muslin cloth.

Sampling of Algae

The water sample was collected from Kempabudi Lake, Chamarajpet around the month of August 2018.

Isolation and growth of algae

The collected sample was inoculated into Algae Culture Broth. After a significant amount of growth, the algae mixture was sub cultured on agar plates in the same Algae Culture media.

The microscopic view of the algal mixture showed the following species of algae.

S. no.	ALGAE
1.	*Anabena*
2.	*Nostoc*
3.	*Spirulina*
4.	*Spirogyra*
5.	*Volvox*
6.	*Diatom*

Identification of algae

The algal isolates that were obtained did not represent the whole algae in the collected samples in this study. Some algae need typical media with typical environmental factors to be grown which were different from those utilized in this study (Abedin and Taha, 2008).

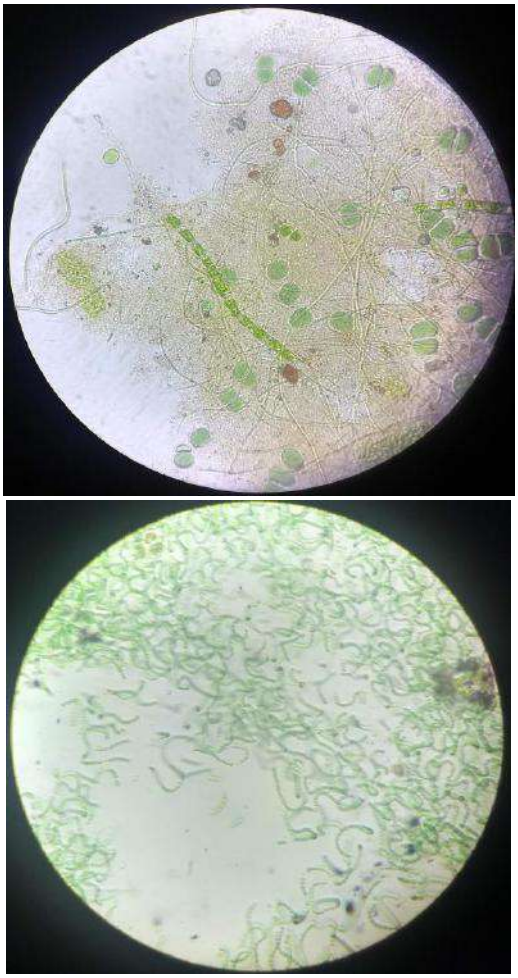


Fig.1: Microscopic views of algae mixture.

https://dx.doi.org/10.22161/ijeab.51.29 *ISSN: 2456-1878*

Control of contaminants

Growth of contaminating bacteria and fungi were prevented by maintaining the pH of the media at 8 - 8.5.

Extraction of *Tamarindusindica*pulp

The fruit of the *Tamarindusindica* was collected from a nearby botanical garden and was sun dried. The dried pulp was administered into the Soxhlet apparatus and the extract was obtained using solvent extraction method by using ethanol.

The extract was subjected to drying to evaporate the ethanol.

Evaluation of the inhibitory effect of the pulp extract

Wells were cut into the algae cultured plate and the extract was administered into the wells. Inhibitory effects of *Tamarindusindica* pulp extract was tested against the mixture of algae depending on the diameters (mm) of inhibition zones through the agar-well diffusion method.

Extraction of *Azadirachtaindica* leaves

The leaves of the *Azadirachtaindica* were collected from a nearby botanical garden and were sun-dried. The dried leaves were administered into the Soxhlet apparatus and the extract was obtained using solvent extraction method by using ethanol.

The extract was subjected to drying to evaporate the ethanol.

Evaluation of the inhibitory effect of the leaf extract

Wells were cut into the algae cultured plate and the extract was administered into the wells. Inhibitory effects of *Azadirachtaindica* leaf extract was tested against the mixture of algae depending on the diameters (mm) of inhibition zones through the agar-well diffusion method.

III. RESULTS

The zone of inhibition was observed on day 2 of administration. The diameter of the zone of inhibition continued to increase until the fifth day. After the fifth day, there was no increase in the diameter. On reducing the concentration of the extract administered, the diameter decreased in direct proportion. On comparison with *Azadirachtaindica* leaf extract, the diameter (in mm) observed was larger for *Azadirachtaindica* than *Tamarindusindica*.

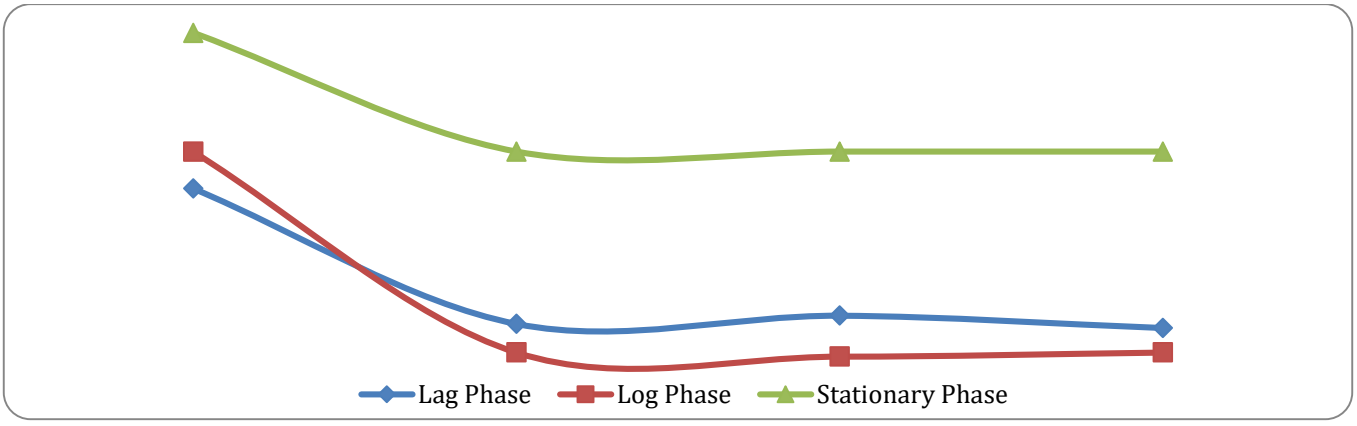


Fig. 2: Growth curve of algae in broth media.

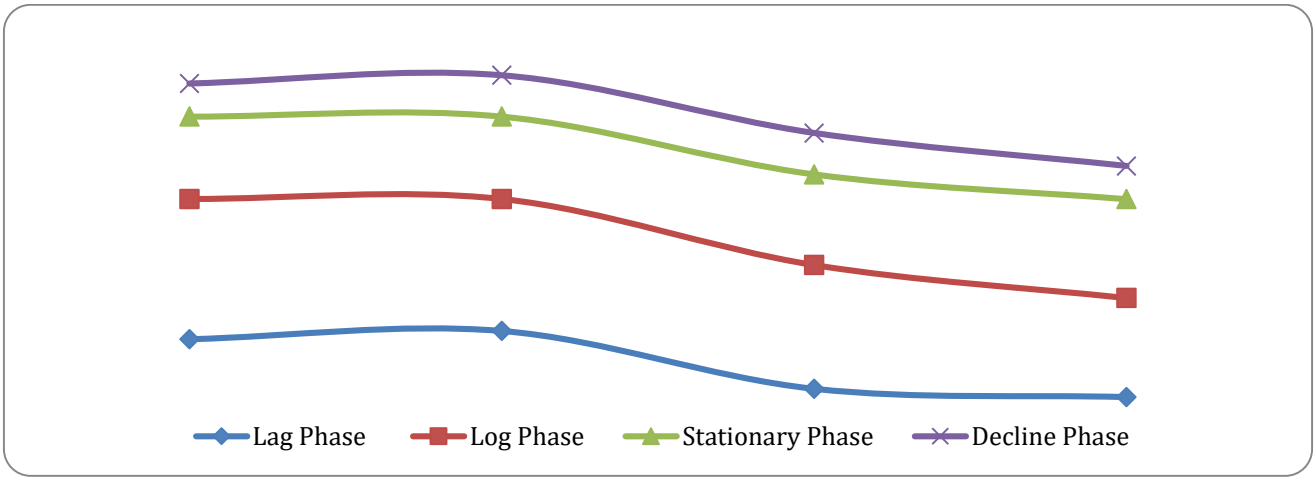


Fig. 3: Growth curve of algae in solid media.

https://dx.doi.org/10.22161/ijeab.51.29 *ISSN: 2456-1878*

Fig.4: Inhibition zone shown by Azadirachtaindica, Tamarindusindica and ethanol on algal culture plate.

Table 1: Indicating zone of inhibition with 100%crude extract of Azadirachtaindica

Concentration	**Culture Plate**	**Volume (µL)**	**Days**	**Diameter of zone of inhibition by *Azadirachtaindica* extract(mm)**
100% Crude extract	1.	75	1	0
			2	12
			3	12
			4	13
			5	13

Table 2: Indicating zone of inhibition with 100%crude extract of Tamarindusindica

Concentration	**Culture Plate**	**Volume (µL)**	**Days**	**Diameter of zone of inhibition by *Tamarindusindica* extract(mm)**
100% Crude extract	1.	75	1	0
			2	11
			3	11
			4	12
			5	12

https://dx.doi.org/10.22161/ijeab.51.29 *ISSN: 2456-1878*

Table 3: Indicating zone of inhibition with 100% absolute ethanol

Concentration	**Culture Plate**	**Volume (µL)**	**Days**	**Diameter of zone of inhibition by Ethanol(mm)**
Absolute ethanol	1.	75	1	0
			2	10
			3	10
			4	11
			5	11

Table 4: Indicating zone of inhibition with 75%crude extract of Azadirachtaindica

Concentration	**Culture Plate**	**Volume (µL)**	**Days**	**Diameter of zone of inhibition by***Azadirachtaindica***extract (mm)**
75% (75 µL of extract + 25 µL DW)	2.	75	1	0
			2	9
			3	9
			4	9.1
			5	9.3

Table 5: Indicating zone of inhibition with 75%crude extract of Tamarindusindica

Concentration	**Culture Plate**	**Volume (µL)**	**Days**	**Diameter of zone of inhibition by***Tamarindusindica***extract (mm)**
75% (75 µL of extract + 25 µL DW)	2	75	1	0
			2	8
			3	8.1
			4	8.4
			5	8.5

Table 6: Indicating zone of inhibition with 75% absolute ethanol

Concentration	**Culture Plate**	**Volume (µL)**	**Days**	**Diameter of zone of inhibition by Ethanol (mm)**
75% (75 µL Absolute ethanol + 25 µL DW)	2.	75	1	0
			2	7
			3	7.2
			4	7.2
			5	7.3

IV. DISCUSSION

This study brought out that there is an initial lag phase in algal growth; the duration of the lag phase decreased on further sub-culturing. It was also noted that algae grow only when the substratum is not completely solid (less agar content) along with the presence of moisture.

Tamarindusindica is known to exhibit antimicrobial activity as it contains flavonoids, alkaloids and polyphenols (Arranz*et al*. 2010). Alkaloids are shown to possess some level of allelopathy on plants (Macias *et al*. 2007).

This study showed that *Tamarindusindica* extracts effectively inhibited the growth of algae. In the case of the 100% crude extract, it has been observed that the zone of inhibition was 11 mm in diameter) on day 2. The magnitude of the zone of inhibition increased on the fourth day (12 mm) and was found to remain constant after that. In the case of 75% of crude extract, the diameter of the inhibition zone was found to be 8 mm on day 2 and the magnitude increased successively up to day 5 (8.5 mm). The diameter of inhibition zones increased with the increased extract concentration. Research has shown that *Azadirachtaindica* has the ability to prevent algal growth (Chia *et al*. 2016).

In contrast with *Azadirachtaindica* leaf extract, the diameter (in mm) observed was larger in case of *Azadirachtaindica*. The zone of inhibition with 100% of crude leaf extract of *Azadirachtaindica* was 12 mm on day 2, while in case of *Tamarindusindica* it was 11 mm. The zone of inhibition with 75% of crude leaf extract of *Azadirachtaindica* was 9 mm on day 2, while in case of *Tamarindusindica* it was 8 mm. The inhibition zone in case of both remained constant after day 5.

Ethanol has been used as a control sample to eliminate experimental error (since the vaporization of ethanol hasn't been done in an ideal method). Ethanol has exhibited a smaller inhibition zone compared to *Azadirachtaindica* and *Tamarindusindica*. The diameter of the zone of inhibition with absolute ethanol was found to be 10 mm on day 2 and increased to 11 mm on day 5. The diameter of zone of inhibition with 75% ethanol was to be 7 mm on day 2 and 7.3 on day 5.

The observed result suggests that fruit extract of *Tamarindusindica*can be used as an alternative way to prevent algal bloom.

V. CONCLUSION

Solvent extract of *Tamarindusindica* is a cheap and effective alternative for the prevention of excessive algal growth which is causing disruption in the aquatic ecosystem. Its inhibitory effect was observed on day 2 and increased upto day 5. On comparison, the leaf extract of *Azadirachtaindica* proved to be more effective than *Tamarindusindica* fruit extract. The comparison was done because both are excellent medicinal plants with effective secondary metabolites. Even without the isolation of specific secondary metabolite of the *Tamarindusindica*fruit, the solvent extract proved to be very efficient. More efficiency of inhibition of algal growth might be achieved by scrutinizing the secondary metabolites of *Tamarindusindica*and administering them specifically. Further studies and investigation need to be done on the effects of the extract on other organisms.

ACKNOWLEDGEMENT

This project would not have been possible without the support of our parents, teachers and friends. We extend our heartfelt gratitude to the Research and Development Cell for providing us with this opportunity to do research at the undergraduate level.

We are deeply and sincerely grateful to our research guide Prof. Mathews P Raj for his constant and immense support and guidance throughout our work. It is our honour to thank Dr. Asha Rajiv, Director, Jain University, School of Sciences for giving us the wonderful opportunity to conduct research and for her constant support and help. We would like to thank Prof. Anitha A Abraham, Head of Department of Life Sciences for her constant advice and facilitating with the equipment and infrastructural support we needed to complete our work.

We extend our gratitude to all the faculty members of the Jain University School of Sciences for being supportive. We are very thankful to the non-teaching lab expertise for assisting and helping us in all the possible ways.

Finally, we thank the administration and management of the Jain University for the financial and infrastructural support.

REFERENCES

[1] Abedin, R.M.A. and H.M.T. Taha. 2008. Antibacterial and antifungal activity of Cyanobacteria and green microalgae. Evaluation of medium components by Plackett- Burman design for antimicrobial activity of Spirulina platensis. Gobal J. Biotech. andBiochem. 3(1): 22- 31.

[2] Ali M. Najem, Abdul Latif M. Jawad and Ayyad W. Raof. 2011. Antagonistic activity evaluation of *Eucalyptus camaldulensis* essential oil in control of algae and associated microorganisms. International Journal of Scientific & Engineering Research Volume 2, Issue 9.

[3] Clark D, Durner J, Navarre DA, Klessig DF. 2000. Nitric oxide inhibition of tobacco catalase and ascorbate peroxidase. Molecular Plant-Microbe Interactions 13: 1380-1384.

[4] Ghorbanian M, Razzaghi-Abyaneh M, Allameh A, Shams-Ghahfarokhi M, Qorbani M. 2008. Study on the effect of neem (*Azadirachtaindica*) leaf extract on the growth of *Aspergillus parasiticus* and production of aflatoxins by it at different incubation times. Mycoses 51: 35-39.

[5] Jianfang Hu, Gan Zhang, Kechang Li, Ping'an Peng, Allan R.Chivas. 2008. Increased eutrophication offshore Hong Kong, China during the past 75 years: Evidence from high-resolution sedimentary records. Marine Chemistry. Volume 110, Issues 1–2, Pages 7-17.

[6] Julio César Escalona-Arranz, Renato Péres-Roses,

https://dx.doi.org/10.22161/ijeab.51.29 *ISSN: 2456-1878*

ImilciUrdaneta-Laffita, Miladis Isabel Camacho-Pozo,1 Jesús Rodríguez-Amado, and Irina Licea-Jiménez. 2010. Antimicrobial activity of extracts from Tamarindusindica L. leaves. Pharmacogn Mag. 242–247.

[7] Macias FA, Molinillo MGJ, Varela RM, Galindo JCG. 2007. Allelopathy a natural alternative for weed control. Pest Management Science 63: 327-348.

[8] Mathias Ahii Chia, JamiuTaiwo Akinsanmi, YahuzaTanimu and ZakariLadan. 2016. Algicidal effects of aqueous leaf extracts of neem (*Azadirachtaindica*) on *Scenedesmusquadricauda* (Turp.) de Brébission.

[9] Okoh OO, *et al*. 2017. Ethanol extract and chromatographic fractions of Tamarindusindica stem bark inhibits Newcastle disease virus replication.

[10] Paerl HW (2006). Assessing and Managing Nutrient Enhanced Eutrophication in Estuaries and Coastal Waters: Interactive Effect of Human and Climate Perturbation. EcolEng 26:40-54.

[11] Paerl HW, Fulton RS, Moisander PH, Dyble J. 2001. Harmful freshwater algal blooms, with an emphasis on cyanobacteria. The Scientific World 1: 76-113.

[12] Paerl HW, Huisman J. 2009. Climate change: a catalyst for global expansion of harmful cyanobacterial blooms. Environmental Microbiology Reports 1: 27-37.

[13] Qiao W, Li C, Fan LM. 2014. Cross-talk between nitric oxide and hydrogen peroxide in plant responses to abiotic stresses. Environmental and Experimental Botany 100: 84-93.

[14] R. John Wallace. 2004. Antimicrobial properties of plant secondary metabolites. Volume 63, Issue 4, pp. 621-629.

[15] Stewart FS, Beswick TS (1979). Bacteriology, Virology and Immunity. 10th Edu, the English Language Book Society and BallierieTindall, London. pp. 35-37.

[16] Tesson B *et al*. 2014. Brown algal morphogenesis: atomic force microscopy as a tool to study the role of mechanical forces. Front Plant Science.

[17] Venugopal R, Liu RH (2012) Phytochemicals in diets for breast cancer prevention: The importance of resveratrol and ursolic acid. Food SciHumWellness 1: 1-13.

[18] United Nations Environment Programm, *The Impact of Eutrophication*, Encyclopedia of Lakes and Reservoirs,2012 Edition.

[19] United Nations, European Commission, International Monetary Fund, Organisation for Economic Co-operation and Development, World Bank, 2005, Handbook of National Accounting: Integrated Environmental and Economic Accounting 2003, Studies in Methods, Series F, No.61, Rev.1, Glossary, United Nations, New York, para. 3.28.

[20] Zhao Q, *et al*. 2018. Temporal and spatial characteristics of potential energy anomaly in Lake Tihu.

[21] Zhang W, Xiong B, Chen L. 2013. Toxicity assessment of *Chlorella vulgaris* and *Chlorella protothecoides*following exposure to Pb (II). Environmental Toxicology and Pharmacology 36: 51-57.

https://dx.doi.org/10.22161/ijeab.51.30 ISSN: 2456-1878

Physicochemical Analysis and Seasonal Variations of Sediment and Water Samples from Selected Surface Waters in Anambra State, Nigeria

Okaa, A .I[1] and Ogu, C.T[2]

Department of Applied Microbiology and Brewing, Nnamdi Azikiwe University , Awka, Nigeria.
Postal Address: Office of the HOD, Department of Applied Microbiology and Brewing, Nnamdi Azikiwe University, Nigeria

Abstract— *The physiochemical properties of sediment and water samples from four surface waters in Anambra State were investigated and their seasonal variations compared. These Physicochemical properties include; temperature, pH , conductivity, colour, total suspended solids, total solids or residues ,total dissolved solids, alkalinity, carbondioxide, total hardness, chloride, Nitrate and Sulphate. Atomic Absorption Spectroscopy was used to determine the mineral elements of the sediment and water. A total number of 24 samples; 3 samples per sampling station (an average of 8 samples) were examined in this study. The average high iron and zinc content ranged from 6.4-52.55 and 0.70-10.7 respectively, which were greater than the World Health Organization permissible range for drinking water. Other physiochemical seasonal differences observed fell within recommended ranges. Many physicochemical properties (Temperature, Conductivity, TSS, TS, Zn, Cu, Fe) of the sampling stations of the water and sediment increased within the rainy season. This showed that increased rainfall, subsequent soil erosion and surface runoff during the rainy season increased the concentration of ions in surface waters. The 't' test analysis showed a significant difference between the average alkalinity values of the rainy and dry season of the water and sediment.*

Keywords— ***Physicochemical, Seasonal variation, Sediment, Surface-water, Water.***

I. INTRODUCTION

Water occupies about 70% of the earth's surface and yet it is one of the scarcest commodities especially in the developing countries of the world. It is one of the most demanded of all urban and rural amenities and it is indispensable for man's activities. Water needs have had serious socio-economic and health influences on urban environment in developing countries where population concentrations have put serious strains on available resources. Amongst the serious environmental problems are waste accumulation and lack of adequate and safe water supply (Buor, 2003; Orji , 2006).

Surface water is a natural water source which collect from water running across the surface of the ground. As this water runs across the ground surface, it picks up microorganisms, organic matter and mineral. Sediment is matter (sand, dirt, gravel) that settles to the bottom of a water. Surface waters in Anambra state serve various purposes ranging from drinking, sources of fish, irrigation to recreation but frequently these waters are polluted. Water pollution is one of the most important environmental problems faced by third world countries (Barry,2000)

The key to effective environmental quality management is the ability to continuously monitor the concentration of various pollutants in the sample of interest. The significance of various substances in water is obvious and it is their level that gives measure of the quality of the water.

II. LITERATURE REVIEW

Water is literally the source of life on earth and is indispensable to man as it is required for both domestic and industrial processes. Chukwura, (2001) stated that because of the numerous uses of water, our bodies of water contain substances introduced directly or indirectly by man, his activities, climate, geophysical or geochemical phenomena whose presence in the water is of such quantity that the quality of the water is impaired or rendered offensive to life hence water pollution results. Effluents which are discharged into the rivers, have increased substantially over the years due to

https://dx.doi.org/10.22161/ijeab.51.30 *ISSN: 2456-1878*

industrialization. Oyeyiola *et al* (2006) stated that the majority of the compounds released into the water have affinity for particulate matter, therefore the chemical composition of bottom sediments reflects the input of discharged substances to the marine environment. Eutrophication is any increase in the concentration of available nutrients; it may be man-made as in sewage discharge into stream or natural as with rain water washings (Chukwura, 2001).

Eutrophication is any increase in the concentration of available nutrients; it may be man-made as in sewage discharge into stream or natural as with rain water washings (Chukwura, 2001). Accumulation of contaminants in the sediments can be linked to local point sources while sediments in more remote areas reflect the overall level of contamination. Gibbs (1993) pointed out that metals of detectable concentrations are found in the environment; the presence of metals in sediments is unavoidable and at low concentrations, metals play an essential role in many biochemical processes. However, they can be deleterious to living organisms at higher concentrations. Angelidis and Aloupi (1995) also noted that metal inputs are subject to a variety of processes that determine their fate and comparison of metal concentrations in sediments or even water, must take into account the processes likely to affect them such as pH and organic matter content of sediments.

Physiochemical analysis is based on actual measurement of physical parameters such as pH, temperature, colour etc and chemical parameters such as metal copper, lead, iron etc and chemical parameters such as metals copper, lead, iron etc. The physiochemical monitoring provides quantitative information about the presence of pollutants in natural streams.. The World Health Organization (WHO) approved chemical standard for potable water as listed in Table 1 below;

World Health Organization Approved Standard (chemical) for portable water

Parameter		WHO limit
pH at 250C		6.5 – 8.5
Chloride (ppm)	250	
Copper (ppm)		1.0
Nitrate (ppm)		4.0
Manganese (ppm)		0.1 – 0.5
Phosphate (ppm)	10	
Sulphate (ppm)	250	
Iron(ppm)		0.3
Zinc (ppm)		3.0
Hardness (CaCO3)(ppm)	200	

(WHO, 2004)

2.1 pH Value: The pH value or hydrogen ion concentration which is a measure of the acidity or alkalinity of a sample, is one of the most important parameters in water chemistry since many of the processes involved in water treatment are pH dependent (APHA, 1980). The pH values of unpolluted water is mainly determined by the interrelationship between free carbondioxide and the amounts of carbonate and bicarbonate present. Thus, the pH values of the most natural waters are in the range of 4 to 9.

2.2 Colour: Colour in water may result from the presence of natural metallic ions, humus and peat materials, weeds and industrial wastes. Water often appear coloured because of material in suspension, so true colour can only be determined after acceptable pre-treatment such as filtration (Chukwura, 2001).

2.3 Alkalinity: Alkalinity is almost entirely due to bicarbonate, carbonate and hydroxide ions in water, usually in association with calcium, magnesium, sodium and potassium ions. The alkalinity of a water resource is its quantitative capacity to react with a strong acid to a designated pH.

2.4 Total Solids/Residue: Total solids refer to solid matter suspended or dissolved in water or waste water. Highly mineralized water also are unsuitable for many industrial application and for these reason, a limit of 500mg residue/litre is desirable for drinking water (Lawal, 1988).

2.5 Suspended solids: Suspended matters are major carriers of many organic and inorganic pollutants including most toxic heavy metals, pathogens and nutrients. Suspended solid is also known as filtratrable solid because it can be filtered.

2.6 Dissolved solids: Dissolved solid is also known as non-filterable solid because it passes through the filter paper. For a given water, the dissolved solids concentration can be directly related to the conductivity. Dissolved solids is obtained as the difference between total solid and suspended solid in a particular quantity of water sample

2.7 Hardness: Water hardness was originally understood to be a measure of the capacity of water to precipitate soap but in conformity to current practice, Twort *et al* (1986) defined total hardness as the sum of the calcium and magnesium carbonate in milligramme per litre.

2.8 Carbondioxide: This is one of the components of the carbonate equilibrium in water. The free carbondioxide content of a water depends on the alkalinity and can contribute significantly to the corrosive properties of water. According to Twort *et al* (1986), surface waters usually contain less than 10mg/I free carbondioxide while some ground waters may easily exceed that concentration.

2.9 Chloride: Chloride ion is one of the inorganic anions in water. In potable water, the salty taste produced by chloride concentration is variable and dependent on the chemical composition of water. Also, a high chloride content may harm metallic pipes and structures as well as growing plants.

2.10 Sulphate: The concentration of sulphate in natural waters can vary over a wide range from a few mg/l to several thousand mg/l. Sulphate can come from several sources such as dissolution of gypsum and the other mineral deposits containing sulphate, oxidation of sulphites and from industrial effluents where sulphates has been used in the manufacturing processes. Sulphurous fuel gasas discharged to the atmosphere in industrial areas often result in acidic rain containing appreciable levels of sulphates.

2.11Nitrate: surface waters, unless badly polluted with sewage effluents seldom contain myuch nitrate. Nitrate is the final oxidation of ammonia and most of the oxidation in soil and water is achieved by nitrifying bacteria and can only occur in well oxygenated environment. Nitrates discharging into receiving water under proper environmental condition degrades stream quality by encouraging excessive growth of algae.

2.12 Metals: the effect of metals in water and waste water range from beneficial through troublesome to dangerously toxic depending on their concentratins. Some metals are essential, others may adversely affect water consumers, waste treatment systems and receiving waters.

III. MATERIALS AND METHODS

3.1 Sample sources and collection

Eight samples on average (4 of water and 4 of sediment) were collected from the following locations; Obizi sediment, Obizi water, Nkisi sediment, Nkisi water, Ebenebe river sediment, Ebenebe river water, Agulu lake sediment, Agulu lake water. Each sample for physiochemical analysis was collected using a clean 2-litre plastic container with screw cap. At the point of collection, the container was rinsed with the sample.

3.2 Physiochemical analysis

Temperature and pH Determination: The temperature and pH of the samples were determined with pH/ temperature metre with reference electrode HANNA MODEL HI, 991001.

3.3 Conductivity determination

The conductivity of the water samples was determined with a DIONIC conductivity meter series 3. The test electrode of the metre was immersed into the water sample and the range was taken by turning the testing range selector gradually until sufficient deflection of the pointer was attained.

Test value = Reading x Factor

3.4 Colour Determination

The standard Lovibond Nessleriser disc (BDH, MK) was used to determine the colour of the samples. The unit adopted in this method of measurement is the colour produced by 1 milligram of platinium per litre of water, which is equivalent to 1 ppm of platinium.

3.5 Total Suspended Solids

Whatman filter paper N0 1001150 was dried, weighed anf fitted into a funnel. Hundrd ml of the sample was filtered through it. The filter paper was removed and dried in the oven at 1050C for one hour. It was later allowed to cool and reweighed (Mamta, 1999)

3.6 Total Solids (Residue)

Clean dry evaporating dishes were ignited at 1050C for one hour in an oven. They were allowed to cool, weighed and kept. Hundred ml of each sample was measured and transferred into each of the pre-weighed dishes and were evaporated to dryness in an oven. It was allowed to cool and then reweighed (Pandey and Carney, 1989)

3.7 Total dissolved solids

The total dissolved solids is easily obtained by simple calculation as followed: total dissolved solids = Total solids – Suspended solids.

3.8 Alkalinity Determination

The double titration method according to Bassat *et al* (1978) was used.

3.9 Carbondioxide Determination

The neutralization titration method according to APHA (1980) was used. Hundred ml of the sample was pipette into a 250ml conical flask, then drops of phenolphthalein indicator was added and titrated with 0.01N to NaOH to a light pink end point.

3.10 Total hardness determination:
Ethylenediaminetetracetic acid (EDTA) titrimetric method according to APHA (1998) was employed in the determination of the total hardness.
3.11Chloride Determination: The argentometric method described in Lawal, 1998 was used to determine the chloride content of the samples. The principle is that in a neutral or slightly alkaline solution, potassium chromate can indicate the end point of the silver nitration of chloride. Silver chloride is precipitated quantitatively before red silver chromate is formed.
Nitrate Determination-Brucine Colorimetric Method:
3.12 Sulphate determination: The principle is that sulphate ion is precipitated in a hydrochloric acid medium with barium chloride so as to form barium tetraoxosulphate (vi) crystals of uniform size. Light absorbance of the BaSO4 suspension is measured by a transmission photometer and sulphate ion concentration is determined from a standard curve. The absorbance of the standard solution was plotted against their concentrations to obtain a curve from which concentrations of sulphate in the sample was obtained.
3.13 Biochemical Oxygen Demand (BOD) Determination
Biochemical Oxygen demand is an empirical test in which standard laboratory procedures are used to measure the relative oxygen requirements of a water sample.
3.14 Chemical Oxygen Demand (COD) Determination: Chemical oxygen demand is the total oxygen consumed by the chemical oxidation of that portion of organic materials in water which can be oxidized by a strong oxidant.Ten ml of water sample was pipette into 250ml conical flask and the following was added: 5ml potassium dichromate, 15ml concentrated tetraoxosulphate (vi) acid and 40ml of distilled water. Seven drops of phenonthroline ferrous sulphate indicator was added and titration was carried out with 0.025N ferrous ammonium sulphate in the burette. Ten ml of blank was also treated with the same reagents as the sample COD was calculated.
3.15 Determination of Mineral Elements
The mineral elements iron, sodium, calcium, lead, copper and zinc were determined by atomic absorption spectrophotometry according to APHA (1998).
Statistical Analysis
The‘t’ test was used to analyse the rainy and dry season alkalinity values of the water and the sediments

IV. RESULTS

Table 1. Physiochemical properties of the sediment and water (rainy season)

	Sampling Stations							
Parameters	**1a**	**1b**	**2a**	**2b**	**3a**	**3b**	**4a**	**4b**
Temperature(oC) 26	28.4	22	26.1	23.5	27	24	28	
pH	5.1	5.45	5.8	6.47	5.23	5.79	6	6.75
Conductivity(uS/cm)	-	1300	-	1500	-	2500	-	1700
Colour(Pt-Co/L) -	5	-	50	-	25	-	25	
TDS (mg/L)	-	178.2	-	148.6	-	178.3	-	168.4
TSS (mg/L)	-	1.8	-	1.4	-	1.70	-	1.60
TS (mg/L)	386	180	450	150	620	180	420	170
Total Hardness (mg/L)20.5		52	9.6	472	14.7	280	24	124
CL (mg/L)	0.32	4	0.24	10	0.16	2	0.4	2
CO2 content (mg/l)	4.22	14.08	1.48	31.68	6.83	10.56	1.48	8.80
Alkalinity (mg/L)52	140	24	160	28	120	99	120	
SO42- (mg/L)	42.5	0.45	22.5	0.6	3.75	1.15	28.75	0.75
NO3 (mg/L)	67.5	2.7	130	3.7	92.5	4.20	110	2.90
Fe (mg/L)	32.9	6.1	27.4	51.6	48.5	10.30	40.00	13.20
Pb (mg/L)	0.08	0.04	0.16	0.15	0.06	0.10	0.04	0.07
Cu (mg/L)	0.95	5.8	2.2	0.6	3.2	2.10	6.40	2.40
Zn (mg/L)	6.3	0.6	10.6	8	1.9	0.40	5.00	3.41
Ca (mg/L)	3	11.3	0.85	2.1	6.91	7.25	4.62	4.06
Na (mg/L)	4.03	7.01	5.14	3.65	5.32	3.96	3.66	2.90

https://dx.doi.org/10.22161/ijeab.51.30 *ISSN: 2456-1878*

Table 2. Physicochemical properties of the sediment and water (Dry Season)

	Sampling Stations							
Parameters	1a	1b	2a	2b	3a	3b	4a	4b
Temperature(oC) 27	31	24	28	26	30	22	27	
pH	5.00	5.30	6.20	6.50	6.10	6.70	6.42	6.85
Conductivity(uS/cm)	-	900	-	1500	-	1400	-	1300
Colour(Pt-Co/L) -	5	-	25	-	10	-	5	
TDS (mg/L)	-	158.36	-	128.7	-	153.48	-	148.6
TSS (mg/L)	-	1.64	-	1.30	-	1.52	-	1.40
TS (mg/L)	320	160	365	130	410	155	400	150
Total Hardness (mg/L)5.60		248	4.96	420	17.76	348	10.88	332
BOD		1.80		2.10		3.00		3.10
COD		7		12		16.75		38.25
CL (mg/L)	0.25	3.98	0.16	7.95	0.24	49.70	0.16	5.96
CO2 content (mg/l)	1.83	10.56	2.96	17.60	3.10	22.88	0.99	5.28
Alkalinity (mg/L)3.2	20	3.2	120	9.60	60	6.40	20	
SO42- (mg/L)	48.75	0.45	22.50	0.55	6.25	1.10	3.75	0.75
NO3- (mg/L)	70	2.80	125	3.75	88.75	4.10	107.50	3.20
Fe (mg/L)	34.80	6.70	30.20	53.50	45.00	11.10	38.20	14.43
Pb (mg/L)	0.10	0.02	0.12	0.11	0.04	0.08	0.03	0.05
Cu (mg/L)	0.15	6.00	2.50	0.68	3.00	2.50	6.85	3.10
Zn (mg/L)	6.00	0.80	9.73	6.42	2.60	0.30	5.40	3.85
Ca (mg/L)	2.30	10.20	0.52	2.60	6.10	6.50	3.00	3.43
Na (mg/L)	3.85	7.55	3.64	4.20	4.83	3.52	2.78	2.35

Table 3. Average physicochemical properties of sediment and water (rainy and dry season)

	Sampling Stations							
Parameters	1a	1b	2a	2b	3a	3b	4a	4b
Temperature(oC) 26.50	29.70	23	27.05	24.75	28.50	23	27.5	
pH	5.05	5.38	6	6.49	5.67	6.25	6.21	6.80
Conductivity(uS/cm)	-	1100	-	1500	-	1950	-	1500
Colour(Pt-Co/L) -	5	-	37.5	-	17.5	-	15	
TDS (mg/L)	-	168.28	-	138.65	-	165.89	-	158.50
TSS (mg/L)	-	1.72	-	1.35	-	1.61	-	1.50
TS (mg/L)	353	170	407.5	140	515	167.5	410	160
Total Hardness (mg/L)13.05		150	7.28	446	16.23	314	17.44	228
CL (mg/L)	0.28	3.99	0.20	8.98	0.20	51.70	0.28	3.98
CO2 content (mg/l)	3.03	12.32	2.22	24.64	4.97	16.72	1.24	7.04
Alkalinity (mg/L)27.60	80	13.60	140	18.80	90	52.70	70	
SO42- (mg/L)	45.63	0.45	22.50	0.58	5	1.13	16.25	0.75
NO3 (mg/L)	68.75	2.75	127.50	3.73	90.63	4.15	108.75	3.01
Fe (mg/L)	33.85	6.40	28.80	52.55	46.75	10.70	39.10	13.82
Pb (mg/L)	0.09	0.03	0.14	0.13	0.05	0.09	0.035	0.06
Cu (mg/L)	0.05	5.90	2.35	0.64	3.10	2.30	6.63	2.75
Zn (mg/L)	6.15	0.70	10.17	7.21	2.25	0.35	5.20	3.63
Ca (mg/L)	2.65	10.75	0.69	2.35	6.51	6.88	3.81	3.75
Na (mg/L)	3.94	7.28	4.39	3.93	5.08	3.74	3.22	2.63

V. DISCUSSION

The investigation revealed that majority of the inhabitants of the catchment communities depend on these surface waters for their domestic uses. There was a relative increase in temperature of the sampling stations for water in the dry season. The accumulated organic load may lead to increase in absorption of heat with a concomitant increase in temperature at these stations. This explanation is in accordance with observations made by Alabaster and Lloyd (1980). The average water temperature observed from sampling stations lies between 25^0C and 35^oC, that is the temperature of natural waters in the tropics as reported by Alabaster and Lloyd (1980).

The average pH values (water) of the surface waters (rainy and dry season) did not fall within the WHO range of 6.5 to 8.5 and therefore not fit for human consumption.

For hardness, only one of the surface water (Obizi River Water) met the WHO limit of 200mg/L, as the average total hardness (seasonal) of others were above limit.

The study also revealed that the average sulphate and nitrate concentrations of surface waters (water) are partly within the WHO limit. This may be attributed to the steady free flowing of the rivers. The higher average seasonal concentration of zinc from Nkisi river (water) might be due to its location. The eutrophication of the river during rainfall from surrounding industrial sites in Onitsha, (a major nearby city) is a key factor. The average seasonal copper concentrations are higher than the WHO limit of 1mg/L, indicating that most of the surface waters have higher concentrations of copper and therefore not fit for drinking. The average seasonal Lead (heavy metal) concentrations are generally high indicating that these surface waters are contaminated.

Conductivity values were higher during the rainy season. This showed that increased rainfall, subsequent soil erosion and surface run offs during the rainy season increased the concentration of ions in the surface waters. This is in accordance with observations made by Odokuma and Okpokwasili (1997). Camp and Messerve (1974) reported that waters with conductivity values in the range of 750 to 2250us/cm are satisfactory for agricultural purposes (irrigation). The surface waters are good for irrigation since their average conductivity values falls within this range. Other physiological seasonal differences observed here were similar to those previously reported by Vila et al (2000).

REFERENCES

[1] Alabaster.S. and Lloyd, R. (1980). Water Quality Criteria for Freshwater Fish. Butterworths, London .pp. 297.

[2] Angelids, M.O. and Aloupi, M. (1995). Metals in Sediments of Rhodes Habour, Greece. Marine Pollution Bulletin, 3 (4-12): 273-276.

[3] Atlas, R.M. (1997). Biological Oxygen Demand. Principles of Microbiology, 2nd edition. McGraw-Hill Company, New York. Pp.792-793.

[4] APHA, (1980). Methods for the Examination of Water and Wastewater. America Public Health Association.5th edition, USA.pp.230-241,249-256.

[5] APHA, (1998). Standard methods for the examination of water and wastewater. Public Health Association. 20th edition. Baltimore Maryland, USA.pp.I,325.

[6] Barry, H.O. (2000). Towards a clean water for third world cities. *Journal of Environment*, **1**(3): 113-127.

[7] Bashir, B.A. and Adebayo, A.A. (2003). Seasonal Variation in quality and outbreak of water borne diseases in Yola, Nigeria. International Journal of Gender and Health Studies, 1 (1):10-15.

[8] Basset., Denney, R. C., Jeffery, G. H. and Mandham, J.(1978). Vogel's Textbook of quantitative inorganic. William Clowes Publishers, London. pp.325-333,501.

[9] Buor, D. (2003). Water Scarcity and Women's Health in the Kumasi Metropol is, Ghana. *International Journal of Gender and Health Studies*. **1**(1):1-9.

[10] Camp, T. R .and Messerve, R. L. (1974). Water and its Impurities.Hutchinson and Ross Inc.Strondsburg, Pennsylvania. pp.356.

[11] Chukwura, E. I. (2001).Sources and Indicators of Surface and Ground water pollution. Aquatic Microbiology. Otoba Press Ltd., Nkpor-Onitsha. pp.4-6.

[12] Gibbs, R.J.(1993). Metals of the bottom muds in Townsville Harbour, Australia. *Envir. Pollution*. **81**(3):297-300.

[13] Egboka, V.C.E.., Nwankwo, G.I., Orajiaka, I.P. and Ejiofor, A.O. (1989). Principles and problems of environmental pollution of groundwater resources with case examples from developing countries. Environmental Heallth Perspective, 83:9-65.

[14] Lawal, A. T. (1988). Studies of heavy metal pollution of some rivers in Plateau-Jos area. Bulletin of Chemical Society of Nigeria. 6:102-103.

[15] Mamta, T. (1999). Quality Assessment of Water and Waste Water. Lewis Publishers, London.

[16] Orji, M.U.,Ezenwaje, E.E. and Anyaegbunam, B.C. (2006). Spatial appraisal of shallow well water pollution in Awka, Nigeria. *Nigerian Journal of Microbiology*, **20** (3): 1389-1394.

[17] Oyeyiola, A. O. Olayinka, K. O. and Alo, B. I. (2006). Correlation studies of heavy metals concentration with sediment properties of some rivers surrounding the Lagos lagoon. Nigerian Journal of Health and Biomedical Sciences.5(1):118-122.

[18] Pandey, G. N. and Carney, G. C.(1989). Environment Engineering. Teta McGraw-Hill Company Ltd., New Delhi. pp.3, 65-72, 105-115.

[19] Twort, A. C., Law F. M. and Crowley, F.W.(1986). Water supply. Arnold International Student Edition.pp.200-220, 230-239.

[20] Vila, L., Contreras, M., Montecino, V., Pizarro, J. and Adams D. (2000). Rapel: A 30 years temperate reservoir. Eutrophication or contamination? Arkiv Fiir

https://dx.doi.org/10.22161/ijeab.51.30 *ISSN: 2456-1878*

Hydrobiologie Special Issues. Advances in Limnologie 55:31-44.

[21] WHO, (2004). Guidelines for Drinking Water Quality. 3rd edition. World Health Organization. Geneva, Switzerland.

[22] Prescott, L.M., Hardley, J.P. and Klein, D.A. (2002). Measuring Water Quality-BOD. Microbiology. 5th edition McGraw-Hill Company, New York. Pp.657-658.

[23] Rao, P.V. (2004). Monitoring and Analysis. Textbook of Environmental Engineering. Prentice-Hall, New Dehli, India. Pp.214-215.

https://dx.doi.org/10.22161/ijeab.51.31 ISSN: 2456-1878

Xylanases and cellulases biosynthesis by selected fungi in a simple and economic bio system using sugarcane straw

Tania Sila Campioni*, Ana Flávia de Azevedo Carvalho, Franciane Cristina de Figueiredo, Douglas Fernandes da Silva, Pedro de Oliva Neto.

Associated Laboratory of Bioenergy Research Institute (IPBEN), Bioprocess Unit, São Paulo State University (UNESP), Av. Dom Antonio, 2100, Zip code 19806-380, Assis, SP, Brazil.

Abstract— *Sugarcane straw (SS) was used in an economic biosystem to evaluate the production of xylanases and cellulases in submerged fermentation (SmF) by axenic and mixed mode from Trichoderma and Aspergillus species. T. reesei QM9414 axenic culture reached the highest xylanase production (90.2 U/mL) and 0.5 FPU/mL of cellulase activity. The evaluation of agro-industrial residues on fibrolytic enzymes production was performed by a D-optimal design, and revealed the best supplementation of 100% SS, while wheat bran and citric pulp showed lower inductive effects on enzymes production. Also, the scale-up in a stirred tank showed the same yield production profile (xylanase ~ 90 U/mL and celullase 0.6 FPU/mL). Xylanase was characterized by an optimum pH of 5-6 and temperature at 50 ºC, and thermal stability was below 50 ºC. The ion Mn2+ (5 and 10 mM) had a stimulatory effect on xylanase activity. The biobleaching application showed that 30 U/g of xylanases during 15 min decreased Kappa number in 9.37. These results indicate SS as an alternative substrate for fungi fibrolytic enzymes production and the xylanase with low cellulase extract as a potential biobleaching application.*

Keywords— ***sugarcane straw, xylanase, cellulase, axenic and mixed cultures, fibrolytic enzymes.***

I. INTRODUCTION

Due to broadened use of renewable energy sources for biofuels and high-value products production in wordwide, including organic wastes mainly produced by agricultural countries, demand for green technologies has increased replacing the extensive usages of fossil fuels (Ferreira-Leitão et al., 2010; Carpio et al., 2019). Sugarcane cultivation is one of the major agricultural activities in Brazil which produced 620.4 million tons in 2018-2019 (Conab, 2019). During the sugarcane burning harvest system, almost 27 kg of carbon dioxide is released into the atmosphere per ton of sugarcane processed, related to burn (40%), fertilizers (20%) and fossil fuels use (18%), thus this quantity can decrease using no-burning system (Figueiredo et al., 2010). The São Paulo state law number 11.241/ 2002 established that, after 2017, 80% of sugarcane, harvesting should be mechanized and after 2021, no more burning will be permitted in mechanized areas. As a consequence of this new system implantation, almost 15 Mg ha^{-1} dry biomass has been left in the field yearly, mainly SS (sugarcane straw) residue (Hassuani et al., 2005).

Straw represents around one-third of the total primary energy of the sugarcane crop, with a composition very similar to the widely used bagasse, mainly cellulose, hemicellulose and lignin, 30, 30 and 25%, respectively (Leal et al., 2013). The straw residue in the soil range from positive impacts, such as increase in the macrofauna (mainly worms and ants), nutrients recycling, water storage, carbon accumulation, control of soil erosion and weed infestation, to negative impacts, such as increase in pest populations and biomass loss production (Leal et al., 2013; Carvalho et al., 2017). In fact, a research showed that 50% of SS residue in the soil is necessary to improve the yield of sugarcane crop but the other 50% should be recovered to be used in eco-friendly processes (Aquino et al., 2017). Depending on the amount and characteristics, that residue could be collected to produce energy or co-products such as enzymes (Carvalho et al., 2016; Silva et al., 2018), xylitol (Hernández-Pérez et al 2016), and biodegradable products such as cups, and straws (Gankin, 2019).

In addition, the enzyme technology has continuously replaced the traditional chemical processes in many areas, especially fine chemical and pharmaceutical industries (Choi et al., 2015). The global market for industrial enzymes expects to increase from nearly $5.5 billion in 2018 to $7.0 billion in 2023 with a compound annual

growth rate (CAGR) of 4.9% for 2018-2023 (Dewan et al., 2017). The importance of enzyme technology includes the knowledge of fermentation and downstream process, and a high number of available enzymes and applications are developed by the improvement of these technologies (Li et al., 2012). In this sense, the use of agro-industrial residues as carbon source for enzyme biosynthesis by microorganisms, which have potential to decrease the production costs and the final price of enzymes (Salmon et al., 2016; Abdullah et al., 2015). Currently, cellulases represent the third higher industrial enzyme production, and their applications are in cotton, paper recycling, juice extraction, detergent and feed industry (Acharya and Chaudhary, 2012). Other important fibrolytic enzymes are the xylanases, responsible for the hemicellulose hydrolysis. Filamentous fungi produce xylan-degrading enzymes, which is the main interest to industrial purposes due to its low-cost production and the final price of the product as well (Abdullah et al., 2015). Mesophilic fungus as the genera *Aspergillus* and *Trichoderma* have a remarkable importance on xylanases and cellulases improvement production, since they can be cultivated in mixed culture (Ahamed and Vermette 2008; Wen et al., 2005; Dhillon et al., 2011).

Although the efficiency of SS as a feedstock and inducer for cellulase production by some microorganisms were reported to *Streptomyces sp* SLBA-08 (Macedo et al., 2013) and *Trichoderma citrinoviride* (Guerra et al., 2006), in literature there is a lack studies of SS as feedstock for xylanase and cellulase production by *T. reesei*, *Trichoderma harzianum* and *Aspergillus fumigatus* in SmF (submerged fermentation).

In the present study, fibrolytic enzymes production was conducted considering the formulation of the culture medium with SS agro-residue and fungi from *Trichoderma* and *Aspergillus* genera, in axenic and mixed cultures. In addition, the biochemical characterization of the xylanases produced in the best conditions was performed considering biobleaching and future application.

II. MATERIAL AND METHODS

2.1 Microorganisms and substrates

The microorganisms tested in axenic cultures were: *Trichoderma reesei* (Tropical Culture Collection of André Tosello Foundation CCT -2768), *T. reesei* QM9414, *Trichoderma harzianum* N51, *T. harzianum* FS09, *Aspergillus fumigatus* M51 and *A. fumigatus* U2370. These cultures were selected in a previous study as the best producers of fibrolytic enzymes (Carvalho et al., 2015). They were cultured in plates containing 3.9% (w/v) Potato Dextrose Agar (PDA) medium for 7 days at 28 ºC and stored at 4 ºC. Lignocellulosic substrates were used as carbon source in the culture medium. The SS was obtained from Água Bonita Mill, Tarumã-SP, Brazil, pretreated (autoclave at 121 ºC, 15 min, 1 atm), and milled (14 mesh). The citrus pulp (CP) (from Citrovita, Catanduva-SP, Brazil) was milled (14 mesh), and wheat bran (WB) was used without any previous treatment (from Moinho Nacional, Assis-SP, Brazil).

2.2 Selection of microorganisms in axenic and mixed cultures

The axenic and mixed strains were cultivated in Erlenmeyer flasks (250 mL) by SmF containing 80 mL medium (m/v): 3.0% pretreated SS, 0.1% $(NH_4)_2SO_4$, 0.0017% $MgSO_4 \cdot 7H_2O$, 0.1% K_2HPO_4, 0.0028% $ZnSO_4$, 0.1% $NH_4H_2PO_4$, 0.06% KCl, 0.1% yeast extract and 0.1% sucrose at pH 4.5 (Silva et al., 2013). Each fungus spores suspension was prepared by incubating the cultures on PDA plates at 28 ºC for about 10 days, until sufficient sporulation was observed. The spores were harvested using 0.1% Tween 80 solution (v/v) for inoculation purposes (about $1x10^6$ cells/mL). Flasks were inoculated and incubated at 28 ºC, in an orbital shaker at 180 rpm for 360 h. The biomass was separated by 15 min centrifugation at 4 ºC and 2900 x g. The liquid fraction was used as a crude enzymes extract. The binary mixtures of *T. harzianum* FS09 and *A. fumigatus* M51; *T. harzianum* FS09 and *T. reesei* QM9414; *T. reesei* QM9414 and *A. fumigatus* M51; as well as the ternary mixture of *T. harzianum* FS09, *T. reesei* QM9414 and *A. fumigatus* M51; in concentration of spores at $1x10^6$ cells/mL for each one, were combined since they are considered the best xylanase and cellulase producers of this study.

2.3 Formulation of culture medium with mixtures of agro-industrial residues for fibrolytic enzymes production

The SmF of selected microorganism was performed in Erlenmeyer flasks (250 ml, with 80 mL of medium described previously (section 2.2) during 288 h of incubation in a shaker at 28 ºC and 180 rpm. D-Optimal mixture design was performed in order to evaluate the effect of individual substrates and the interactions among them in ternary mixtures on xylanase and cellulase production (Fernández-Núñez et al., 2016; Nunes et al., 2017). The number of experimental combinations in each experimental design was enough to fit special cubic models for response variables. The parameters and restrictions of the mixtures were: SS (60–100% w/w range), CP (0–40% w/w range) and WB (0–20% w/w range). A control experiment using 100% (w/v) of each substrate was performed at the same conditions. The D-

https://dx.doi.org/10.22161/ijeab.51.31 *ISSN: 2456-1878*

optimal experimental design was set up with restrictions and analyzed using Design-Expert software (Design-Expert® software, version 10, Stat-Ease, Inc., Minneapolis, MN, USA). The statistical results were made considering a significance level of 0.05. The strength of linear relationships between actual and predicted values by different models was assessed using the linear correlation coefficient (R^2). The xylanolytic activity in ternary mixtures of agro-industrial residues D-Optimal experimental design was optimized using a desirability function. The optimization criterion was to maximize xylanase activity according to a fitted polynomial for this variable.

2.4 Stirred tank bioreactor culture

The enzyme production by selected microorganism was scaled-up in 2 L BioFlo 115 fermenter (New Brunswick, New Jersey, USA) using medium and inoculation as previously described (section 2.2), working volume of 1.5 L, and Rushton impeller. The culture conditions were 28 °C, 1.7 volume of air per volume of medium per minute (vvm), pH 4.5 for 288 h. Dissolved oxygen was measured by an oxygen electrode and pH was measured and controlled with 1.0% (v/v) H_2SO_4 and 1.0 M NaOH.

2.5 Biochemical characterization of fungal xylanase

The biochemical characterization of xylanases produced from selected microorganism in SmF using selected substrate as described in the following protocols (Carvalho et al., 2006; Carvalho et al., 2010).

2.5.1 Optimum pH and stability

Optimum pH was evaluated by measuring enzyme activity at 50 °C using different buffers: sodium acetate (pH 3.0-6.0), sodium phosphate (pH 6.0-8.0), Tris-HCl (pH 8.0-9.0), and glycine-NaOH (pH 9.0-11.0) and a reaction mixture containing 0.65 mL 0.5% (w/v) xylan in 0.25 M buffer and 0.10 mL crude enzyme. For pH stability, crude enzyme extract was diluted (1:1) in buffers and maintained at 25 °C for 20, 40 and 60 min. An aliquot was used to determine the remaining activity (section 2.6).

2.5.2 Optimum temperature and thermostability

The optimal temperature was determined by incubating the reaction mixture at 20-70 °C (10 min) and assaying the enzyme activity at the optimum pH, in the same reaction mixture (2.6). For thermostability assay, the enzyme solution was incubated at various temperatures (20-70 °C) for 20, 40 and 60 min at pH 5.0 in sealed tubes to prevent evaporation. The enzyme solution was maintained at these temperatures and times. Aliquots were removed and placed on ice before assaying for residual enzyme activity at optimum pH and temperature.

2.5.3 Effect of ions and EDTA

The effects of ions (Cu^{+2}, Mg^{2+}, Mn^{2+}, Zn^{2+}, Fe^{3+}, Ag^{+}) and EDTA (Ethylenediamine tetra-acetic acid) on xylanase activity were evaluated. Solutions concentrations of 5 and 10 mM were added to the reaction mixture at the concentration of 0.2% (v/v). The calculation of the percentage of enzyme activity was performed based on the reference sample without addition of any ion.

2.6 Enzymes activity assay

Xylanase activity was assayed at 50 °C in a reaction with 0.1 mL raw enzyme extract and 0.65 mL of 0.5% (m/v) xylan Birchwood solution (Sigma-Aldrich) in 250 mM sodium acetate buffer, at pH 5 for 10 min (Bailey et al., 1993). The reducing sugar concentration was quantified by the dinitrosalicylic acid (DNS) method (Miller, 1960). One unit (U) of xylanase activity was defined as the amount of enzyme to release 1 µmol of reducing sugar per minute per mL of reaction. The cellulase activity was determined by Ghose (1987). One FPU here is defined as µmoles glucose equivalents released from Whatman nº. 1 per min averaged over 60 min, considering the low enzyme concentration in the raw enzymatic extract.

2.7 Biobleaching

Xylanase from *T. reesei* QM9414 was studied for biobleaching process of Kraft pulp as well as to evaluate its potential use as biobleaching agent. The amount of enzyme used for hydrolysis was 30 units of enzyme per gram of pulp samples. Test conditions were performed in a sealed polyethylene bags with sodium acetate buffer (pH 5.0), at 50 °C for 15 min (soaking stage). Treatment started by diluting the enzyme in the same buffer (pre-heated at 50 °C), adding the solution on pulp samples and then mixed by kneading the bags during 30 s. The final pulp content in the reaction mixture was 3%. Controls were prepared by adding distilled water instead of enzyme. After the enzymatic hydrolysis, the bags were boiled at 100 °C for 5 min to disable the enzymes, cooled and filtered on a Büchner funnel to form paper sheets, used for kappa number analysis.

https://dx.doi.org/10.22161/ijeab.51.31 *ISSN: 2456-1878*

III. RESULTS AND DISCUSSION

3.1 Selection of fungi for fibrolytic enzymes production in axenic and mixed cultures using SS as a carbon source

3.1.1 Axenic fungal cultures

All tested microorganisms showed xylanases and cellulases production using SS substrate as the sole carbon source in SmF (Fig. 1A and 1B). *T. reesei* QM9414 strain stood out compared to other fungi tested, reaching the highest production of 90 U/mL for xylanase and 0.56 FPU/mL for cellulase at 288 h of fermentation. Nevertheless, the fungi *A. fumigatus* M51 and *A. fumigatus* U2370 also showed good results for xylanases production, approximately 70 U/mL (Fig. 1A). However, after 288 h the enzymes activities decreased, probably due to protease presence in SmF (Silva et al., 2016; Haab et al., 1999). In literature, a higher concentration of xylanase was obtained when compared to 3.38 U/mL at 120 h of cultivation by *Trichoderma inhamatum* (Silva et al., 2015). Also, xylanase activity achieved 43.7 U/mL at 144 h of cultivation by *T. reesei* CCT 2768, 35 U/mL by *A. fumigatus* M51 and 28 U/mL by *A. fumigatus* U2370, using sugarcane bagasse in culture medium (Carvalho et al., 2010).

The fungi *T. harzianum* FS09, *A. fumigatus* M51 and *T. reesei* QM 9414 were the best cellulases producers, 0.2, 0.4 and 0.6 FPU/mL at 288 h, respectively (Fig. 1B). However, these results obtained to cellulases were lower compared to those found in other studies such as Zhang et al (2014) (0.93 FPU/mL, 96 h) and Xiong et al (2016) (2.33 FPU/mL, 144 h) also produced by *Trichoderma* species, although in these studies were used different substrates as pretreated corn stover and a synthetic medium, respectively. The fact that *T. reesei* QM 9414 produced low cellulases is important for pulp biobleaching application of xylanases for reducing the chlorinated compounds in the paper mills.

3.1.2 Mixed fungal cultures

The mixed fungal and axenic cultures were compared in the present study. Since the *Trichoderma* and *Aspergillus* co-culture system has been reported in literature (Ahamed and Vermette, 2008; Wen et al., 2005), the followed mixtures were proposed: *T. reesei* QM 9414, *A. fumigatus* M51 and *T. harzianum* FS09. Xylanase and cellulase production profile by mixed cultures during 360 h of cultivation were evaluated (Fig. 2A and 2B).

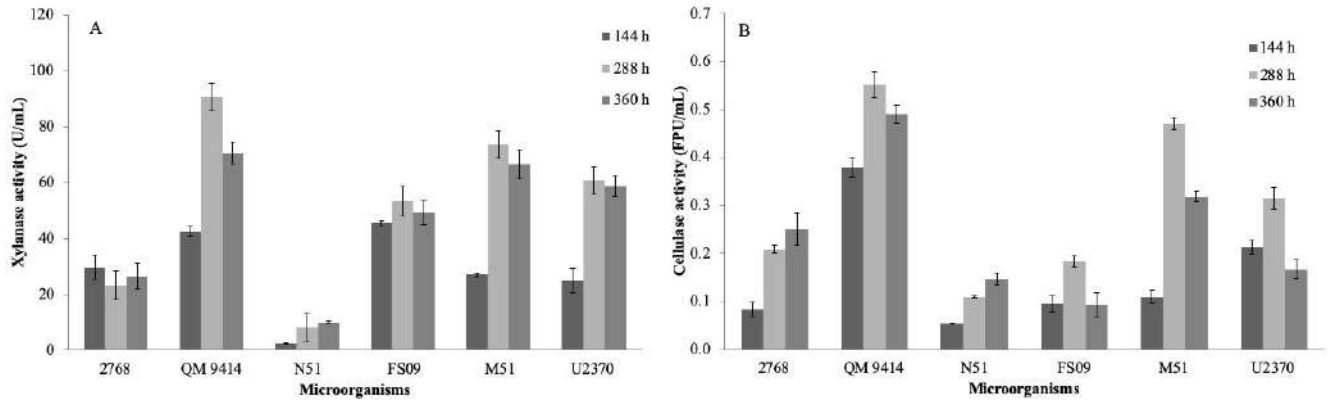


Fig. 1: A) Profile of xylanase production by fungi: T. reesei 2768 (2768), T. reesei QM9414 (QM9414), T. harzianum N51 (N51), T. harzianum FS09 (FS09), A. fumigatus M51 (M51) and A. fumigatus U2370 (U2370), in SmF using SS as substrate (28 ºC, pH 4.5, 180 rpm). Each bar value was the average of three replicate experiments, and the error bars show the data ranges. B) Profile of cellulase production by fungi: T. reesei 2768 (2768), T. reesei QM9414 (QM9414), T. harzianum N51 (N51), T. harzianum FS09 (FS09), A. fumigatus M51 (M51) and A. fumigatus U2370 (U2370), in SmF using SS as substrate (28 ºC, pH 4.5, 180 rpm). Each bar value was the average of three replicate experiments, and the error bars show the data ranges.

When fibrolytic enzymes biosynthesis from these mixed cultures were compared to axenic culture (Fig. 1-2), the enzyme activities of mixed cultures were lower. However, this result was not expected according to literature (Ahamed and Vermette, 2008; Wen et al., 2005; Dhillon et al., 2011), since the mixed cultures with *Trichoderma* and *Aspergillus* genera resulted in a complete enzymatic pool that acts synergistically better in substrate degradation compared to respective axenic culture. According to Duff et al. (1987), fungi species started a substrate competition between them, consequently blocking the enzyme production. The fibrolytic enzymes biosynthesis by *Aspergillus* inhibited the enzymes biosynthesis of *Trichoderma*, probably due

https://dx.doi.org/10.22161/ijeab.51.31 *ISSN: 2456-1878*

to the catalysis of those enzymes already produced. Proteases or endotoxins biosynthesis could degrade or inhibit the cellulases. In addition, a competition between these microorganisms for the same nutrients in the medium is another hypothesis. The carbon source is reported an important parameter to a successful mixed culture (Dhillon et al., 2011).

Although the results were lower than axenic cultivation for xylanase production, the mixed culture *T. reesei* QM 9414 and *A. fumigatus* M51 reached the maximum value of 60 U/mL (Fig. 2A). On the other hand, it was better than produced by Zhang et al. (2014) (2.5 U/mL), but with another strain (*T. reesei* Rut C-30). These authors also reported a slightly improvement on cellulase production (22.89 - 24.17 U/g) respectively from axenic to mixed cultures, in solid state fermentation (SSF), while the substrate consumption was better in mixed culture. *T. reesei* mutant and *A. niger* in mixed culture resulted in an improvement on enzymes production comparing to single culture by non-mutant strain (Gutierrez-Correa et al., 1999). A synergy in mixed culture of *Trichoderma* and *Aspergillus* was also verified for substrate degradation and consequently a higher enzyme synthesis (Ahamed and Vermette, 2008). However, the culture of *T. reesei* and *A. phoenics* ATCC329 xylanase was worse compared to axenic culture in the present study (Wen et al., 2005). Enzymes production by a single culture is preferred to achieve the better substrate degradation from its synergic effect, despite the mixed culture improves cellulases and β-glucosidases production by *T. reesei* QM9414 and *A terreus* SUK-1 (Wen et al., 2005). In fact, other authors reported the competition by *Trichoderma* and *Aspergillus* to the same nutrients in the medium in a mixed culture (Ahamed and Vermette, 2008; Duff et al., 1987; Anthony et al., 2016). As *T. reesei* showed a great production of xylanases, this strain was selected for the next steps of this work with emphasis for xylanases.

For fibrolytic enzymes production, 3% (m/v) of the substrates SS, CP and WB were evaluated isolated by *T. reesei* QM9414 in SmF medium (Table 1). The culture medium formulated by SS only as substrate showed a higher performance for xylanases biosynthesis (90 U/mL) than other residues. For cellulases production, the cultures of *T. reesei* QM9414 also showed a highest preference for SS (0.6 FPU/mL) (Table 1).

Table 1: Fibrolytic enzymes production by T. reesei QM 9414 using agro-industrial residues and its respective chemical composition.

Substrate**	Xylanase activity (U/mL)	Cellulase activity (FPU/mL)	Cellulose (%) w/w	Hemicellulose (%) w/w	Lignin (%) w/w	Reference
Sugarcane Straw	90.6±7.04	0.56±<0.1	33.77	27.38	21.28	Szczerbowski, et al., 2014
Wheat bran	37.7±4.23	<0.10±<0.1	22.3	32	4	Marín et al., 2007
Citrus pulp	31.0±5.87	0.10±<0.1	24.52	7.57	7.51	Rahman et al., 2017

*The results are related with the average and standard deviation of three experiments. **(3% w/v).

https://dx.doi.org/10.22161/ijeab.51.31 *ISSN: 2456-1878*

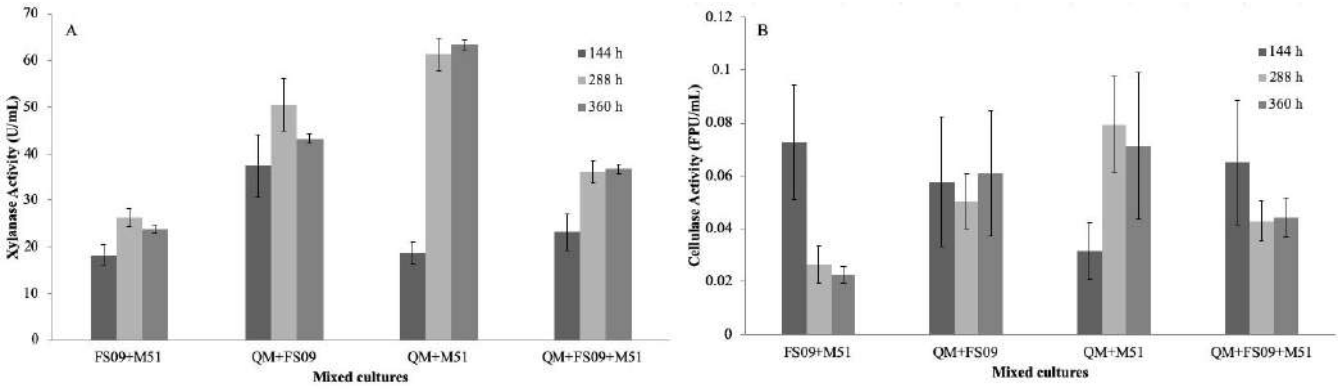


Fig. 2: A) Profile of xylanase production in mixed cultures: T. harzianum FS09 + A. fumigatus M51 (FS09+M51); T. harzianum FS09 + T. reesei QM 9414 (QM+FS09); T. reesei QM 9414+ A. fumigatus M51 (QM+M51); T. harzianum FS09 + T. reesei QM 9414+ A. fumigatus M51 (QM+FS09+M51), in SmF using SS as substrate (28 ºC, pH 4.5, 180 rpm). B) Profile of cellulase production in mixed cultures: T. harzianum FS09 + A. fumigatus M51 (FS09+M51); T. harzianum FS09 + T. reesei QM 9414 (QM+FS09); T. reesei QM 9414+ A. fumigatus M51 (QM+M51); T. harzianum FS09 + T. reesei QM 9414+ A. fumigatus M51 (QM+FS09+M51), in SmF using SS as substrate (28 ºC, pH 4.5, 180 rpm). Each bar value was the average of three replicate experiments, and the error bars show the data ranges.

3.2 The effect of the mixture of agro-industrial residues in formulated media for fibrolytic enzyme production by *T. reesei* QM 9414

The use of WB as substrate was proposed since in literature was observed higher xylanase production in SSF culture (Dhillon et al., 2011; Guimarães et al., 2013) for this residue. The substrates compositions (Table 1) suggest that CP and WB should be more easily hydrolyzed due to their low lignin content. In addition, this fact is responsible for a better xylanases production in SS, since SS residue has high level of lignin makes the degradation of the fiber more difficult and it demands more fibrolytic enzymes.

In the second set of experiments, a D-Optimal mixture experimental design was used to evaluate the synergistic or antagonistic effects of the mixed carbon sources in SmF to produce fibrolytic enzymes by *T. reesei* QM 9414 in 12 days (Table 2). When xylanase and cellulase activities were evaluated, for ternary mixtures of these substrates, were modeled in D-optimal design, cubic models were satisfactorily fitted to the experimental data (model significance tests, $p<0.05$ and lack of fit tests, $p>0.05$).

Xylanase activity (U/mL) = 89.18*A+80.18*B+1408.6*C-3.97*AB-2716.56*AC-2693.27*BC+3926.94*ABC+269.98*AB(A-B)+1683.22*AC(A-C) +1.798*BC(B-C) **Eq. (1)**

Cellulase activity (U/mL) = 0.52*A+0.43*B+10.87*C-0.11*AB-21.73*AC-21.4734*BC+30.88*ABC+1.88*AB(A-B)+11.89*AC(A-C) + 13.45*BC(B-C). **Eq.(2)**

The equations for xylanase and cellulase activities (Equations 1-2 for actual values) in conjunction with contour Graphs (Fig. 3A and 3B) showed the major contribution of SS for higher values of fibrolytic enzymes activities.

The SS influence on xylanase activity was noticed that activity increased with higher substrate concentration, while for CP residue a slight increment on xylanase activity was observed. The substrate WB was not interesting for this purpose since the results were not satisfactory.

Table 2: Results derived from D-optimal experimental design for ternary mixtures of SS, CP and WB as carbon sources in SmF by T. reesei QM9414 (pH 4.5, 28 ºC, 288 h).

Experiment	Sugarcane Straw (% m/m)	Citrus Pulp (% m/m)	Wheat Bran (% m/m)	Xylanase Activity (U/mL)	Cellulase Activity (FPU/mL)
1	80.0	0.0	20.0	69.4	0.3
2	75.0	15.0	10.0	83.9	0.4
3	60.0	20.0	20.0	70.0	0.3

https://dx.doi.org/10.22161/ijeab.51.31 *ISSN: 2456-1878*

4	60.0	40.0	0.0	81.8	0.4
5	60.0	20.0	20.0	71.8	0.3
6	90.0	0.0	10.0	67.3	0.2
7	82.5	7.5	10.0	93.3	0.4
8	66.67	20.0	13.33	61.2	0.2
9	60.0	40.0	0.0	78.4	0.4
10 (C)	100.0	0.0	0.0	**90.2**	**0.5**
11 (C)	100.0	0.0	0.0	88.1	0.5
12	80.0	20.0	0.0	83.9	0.4
13	70.0	10.0	20.0	84.7	0.4
14	67.5	27.5	5.0	88.2	0.4
15	80.0	20.0	0.0	83.5	0.5

The math models are expressed in Eq. 1-2, with coded variables showing the enzymatic activities as function of: A = SS (w/w), B = CP (w/w), and C = WB (w/w). According to ANOVA, each activity response desired, xylanase and cellulase activities produced were statistically significant ($p<0.05$), respectively, for the cubic math models with high Regression coefficient (R^2_{adj}= 0.95, 0.93).

Regarding the cellulase production, SS in a relatively higher concentration presented great activities. However, WB did not represent any synergic effect with other substrates. CP presented a positive effect on cellulase activity within the range interactions. On the other hand, these results are in disagreement with some authors that found an improvement on enzymes production in optimization studies of mixed substrates. Das et al. (2013) showed cellulase production increased 1.3-fold after the medium optimization, containing WB and rice straw by *A. fumigatus* ABK9. WB also performed a positive effect (21%) in the xylanase production by *A. flavus* (Guimarães et al., 2013).

Considering the final purpose of the use of crude enzymatic extract rich in xylanases and poor in cellulases, which are an important characteristic for biobleaching of kraft pulp (Guimarães et al., 2013; Nagar et al., 2010), the optimization of parameters was adjusted to reach a maximum of xylanases and low cellulases production. The optimal set of factors to maximize xylanase production by *T. reesei* was 100% SS, which the experiment 10 reached 90.2 U/mL (Table 2). The most significant results were achieved with 100% SS with desirability predicted for the model was 0.92. The result was validated (in triplicate) in the same conditions (100% SS). The predicted result from the desirability function was 89.2 U/mL and the result obtained, 90.2 Um/mL, presented no significant difference (Anova+Tukey, $p>0.05$). The crude enzymes extract under this condition was rich in xylanases and poor in cellulases, a ratio of 1:0.005 U/mL, respectively.

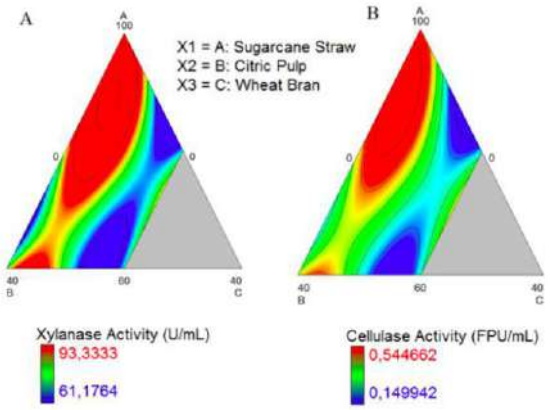


Fig. 3: Contour plots of responses generated by the interactions of the A= SS (w/w); B= CP (w/w); C= WB (w/w), on fibrolytic activities. A) Xylanase activity and B) Cellulase activity produced in SmF by T. reesei QM9414 using SS, CP and WB as substrates (28 ºC, pH 4.5, 180 rpm).

Regarding the xylanase application on kraft pulp biobleaching, Campioni et al. (2019) studied xylanase extract produced by *T. reesei* QM9414 in SmF with SS and optimized the biobleaching parameters. The best conditions were 30 U/g of xylanase, at pH 5, at 50 °C during 30 min and resulted a 12.5% of Kappa number reduction. After the xylanase biobleaching, the final chlorine dioxide consumption reduced to 10%, maintaining the same brightness compared to control on the subsequent chemical process. In addition, an important parameter for biobleaching application is the xylanase combined with low cellulase concentration or

https://dx.doi.org/10.22161/ijeab.51.31 *ISSN: 2456-1878*

even no cellulase activity, otherwise, higher amount of this enzyme could degrade the pulp.

It is known about the successful application of enzymes depends not only on the substrate choice but a simple bioprocess and mainly a low-cost production as well. As mentioned previously (section 1), regarding the transition of no sugarcane burning on harvest system (São Paulo State No. 11.241/2002), the SS residue has been left large amounts on fields, which influenced the dynamics of sugarcane production in several aspects (Carvalho et al., 2017). Additionally, SS has been considered a low-cost residue, which the average of value of US $9.38/ton (Carpio et al., 2019). In this sense, several lignocellulosic agro-industrial residues have been widely evaluated as substrate for xylanase production, such as sugarcane bagasse, WB, sawdust, soy flour, maize straw and others (Knob et al., 2013). Although the use of agro-industrial residues has been extensity described in literature, there is the concern about multiple and complex process steps, consequently become more expensive and difficult to scale up. For example, the substrate pretreatment procedures, waste of extensive washing with distilled water (Knob et al., 2014), chemical pretreatments and in some cases they can generate other toxic compounds for microorganisms and become difficult to find an appropriate destination (Robl et al., 2015). Therefore, this study is a cost effective and simple using SS as a potential substrate for fibrolytic production by *T. reesei* QM9414 and its biobleaching application. After the selection of microorganism and agro-industrial residue used as carbon sources, the enzymatic production was scaled up in bioreactor using 1.5 L working volume and controlled conditions, resulting in 88.02±4.54 U/mL and 0.41±<0.1 FPU/mL, for xylanase and cellulase respectively, proving a high level of xylanase production using 100% of SS by *T. reesei* QM9414 can be obtained by this simple and economical bioprocess. On the other hand, the enzyme production losses were detected in scaling-up of *T. harzianum* P49P11 in SmF using sugarcane bagasse in stirred tank bioreactor (Haab et al., 1990).

3.3 Xylanases biochemical characterization

The enzymatic extract produced by *T. reesei* QM9414 cultivated in SS medium (12 culture days) showed the highest xylanase activity at pH 5 (100 U/mL) (Fig. 4A). The lower range (pH 3-4) and basic pH (pH 8-11) strongly decreased the enzymatic activity. In spite of this, when basic pH was performed the Tris-HCl buffer was chosen than sodium phosphate due to higher enzyme activity in the same pH 8, respectively 65 and 20 U/mL. Xylanase residual activities linearly decreased after the incubation time (20, 40 and 60 min) for all pH ranges (Fig. 4B). The loss of activity varied from 20-95% compared to control, and a higher loss was at pH 8, after 60 min of incubation. In the range of pH 5-6, the enzyme remained 80% active after all incubation times. Xylanases from other *Trichoderma* species was also found in literature with optimum pH 5-6, but with broader pH ranges (Table 3).

Considering pH close to 5.0 as xylanase optimum pH, some applications were found in literature. Zhang et al (2014) proposed the use of xylanases as an additive in bird feed, due to pH range used in this feed was 5.5-6.5. Other sectors are possible such as juice mills (Nagar et al., 2010) and bioethanol (Ferreira-Leitão et al., 2010; Carpio et al., 2019).

In this study, xylanase *T. reesei* QM9414 optimum activity was observed at 50 ºC (Fig. 5A). This temperature is commonly reported by *Trichoderma SC9* and *T. inhamatum* (Tab. 3), beyond microorganisms from other genera: *Paenibacillus macquariensis* (Terrasan et al., 2013) and *Penicillium janczewskii* (Jänis et al., 2001).

The Fig. 5B depicts the thermostability. In temperatures of 20-30 ºC, and after 20, 40 and 60 min, xylanase retained almost 80% of its activity. On the other hand, in temperatures higher than 50 ºC, a linear decrease in enzymatic activity was observed, except at the point at 50 ºC for 20 min, which the activity just improved slightly and then decreased again. Xylanase produced by *T. reesei* QM9414 showed optimum at 50 ºC temperature of incubation. The low thermostability of xylanase by other species of *Trichoderma* was also observed in literature.

https://dx.doi.org/10.22161/ijeab.51.31 *ISSN: 2456-1878*

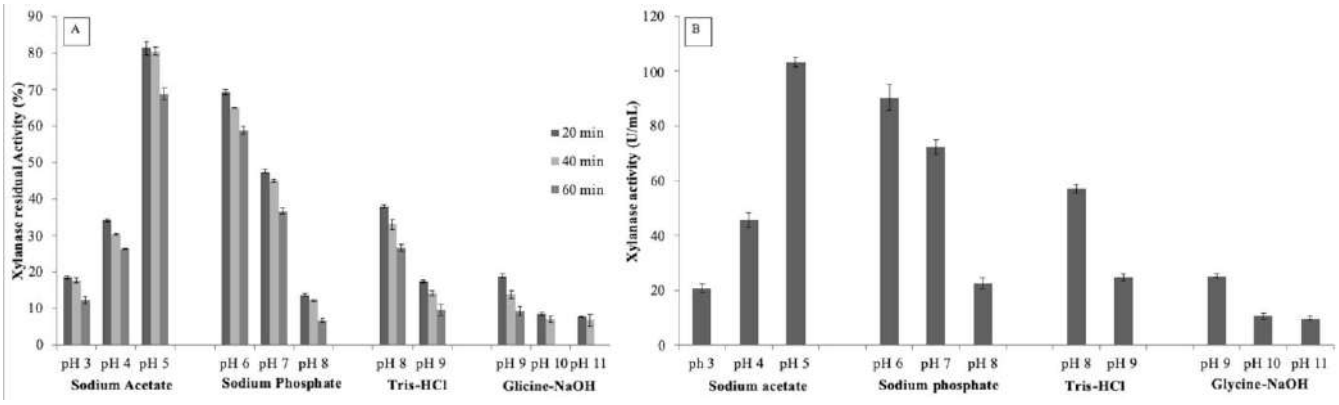


Fig. 4: A) Effect of pH on xylanase activity of the crude extract produced by T. reesei QM9414 cultivated with SS (pH 4.5, 28 ºC, 288 h). Each bar value was the average of three replicate experiments, and the error bars show the data ranges. B) Effect of pH on xylanase activity stability of the crude extract produced by T. reesei QM9414 cultivated with SS (pH 4.5, 28 ºC, 288 h). Each bar value was the average of three replicate experiments, and the error bars show the data ranges.

Table 3: Comparative xylanase characteristics produced by different Trichoderma species in literature.

Microorganism	Optimum pH	Stability range pH	Optimum temperature (ºC)	Reference
T. reesei QM9414	**5.0**	**5.0-6.0**	**50**	**This work**
T. inhamatum	Xyl I: 5-5.5 Xyl II: 5	Xyl I: 4.5-6.5 Xyl II: 5.0	50 (both)	Silva et al., 2015
Trichoderma sp SC9	6.0	3.5-9.0	42.5	Zhou et al., 2011
T. harzianum 1073 D3	5.0	3.0-7.0	60	Isil and Nilufer, 2005
T. reesei	6.0	3.0-8.0	-	He et al., 2009

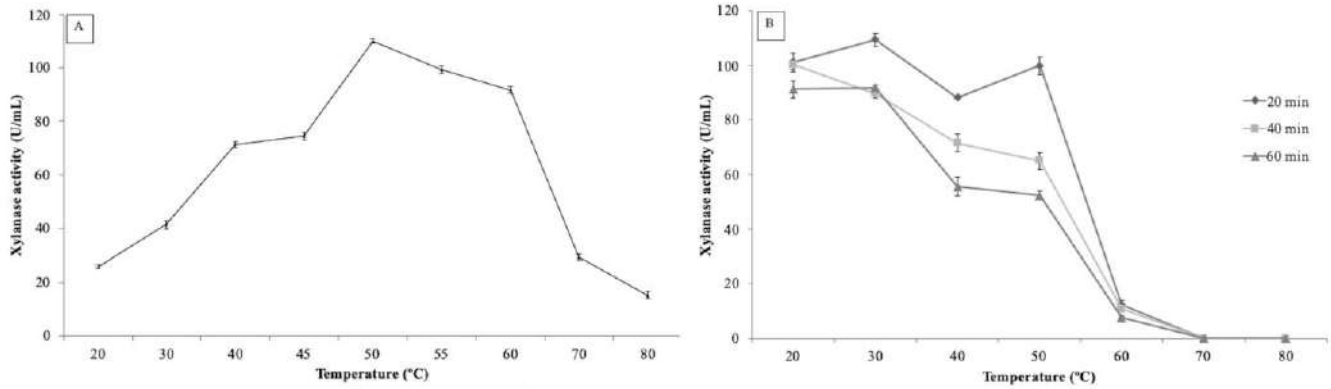


Fig. 5: A) Effect of temperature on xylanase activity produced from T. reesei QM9414. B) Thermostability of xylanase produced by T. reesei QM9414 (pH 4.5, 28 °C, 288 h). Each bar was the average of three replicate experiments, and the error bars show the data ranges.

The thermostability of *T. inhamatum* xylanase presented a half-life of 2.2 h at 40 ºC, and subsequently when the temperature reached 50 ºC this time dropped drastically to 2 min (Silva et al., 2015). Another work showed the stability of *T. reesei* RUT C-30 xylanase was 94% at 50 ºC after 30 min of incubation (He et al., 2009). The thermostability loss of xylanase from *Trichoderma* genus in temperatures higher than 50 ºC can be explained by a conformational structure change (Lopéz and Estrada, 2014), as well as the loss of secondary structure at 58.8 ºC and tertiary one in 56.3 ºC, reflecting in decrease of activity (Cobos and Estrada, 2003). Some additives in xylanases can be applied to solve the thermostability loss, such as polyhydroxylic co-solvents addiction (Xiong et al., 2004) and mutations in bisulfide bounds (Blanco et al., 1995). The effect of activation or inhibition of ions

https://dx.doi.org/10.22161/ijeab.51.31 *ISSN: 2456-1878*

and EDTA on xylanases activities were evaluated and considering two ions solution concentrations, 5 and 10 mM. When the Cu^{2+}, Mg^{2+}, Mn^{2+} and Zn^{2+} ions were added, there was an increment on the enzymatic activity (Table 4). The most expressive result was the Mn^{2+}, 39 and 49%, for the respective concentrations. In contrast, 10 mM of ions Cu^{2+} and Ag^{+} resulted in a strong inhibition of xylanase, 21 and 18% respectively. In literature, the presence of Mn^{2+} and Zn^{2+} also increased xylanase activity produced by *T. harzianum* 1073 D3, whereas in the presence of Mg^{2+} and Cu^{2+} the activity was not affected (Isil and Nilufer, 2005). According to Blanco et al. (1995) Mn^{2+} and Cu^{2+} did not affect the xylanase activity, while Mg^{2+} had a stimulatory effect. In addition, Mn^{2+} also stimulated the enzymatic activity for xylanases from *Paenibacillus macquariensis* (Terrasan et al., 2013). In this last work Cu^{2+} and Fe^{3+} caused inhibition on the enzymatic activity, whereas Mn^{2+} and Mg^{2+} presented no difference compared to control. EDTA caused a slightly decreased on the xylanase activity at concentrations of 5 and 10 mM, 10 and 0.8%, respectively (Table 4). The explanation of the authors for this fact was that an enzyme needs divalent ions for catalysis. In other works, EDTA caused inhibition of the enzymatic activity of xylanases in the concentrations of 1, 2 and 10 mM (Silva et al., 2008).

3.4 Bioblеaching

In order to evaluate the xylanase efficiency for cellulose pulp biobleaching, the pulp was clarified by *T. reesei* QM9414 crude extract and 30 Units of xylanase per gram of pulp in 15 min was successfully effective compared to controls. Xylanase reduced the kappa number in 9.37% (2.1 kappa points). In literature, xylanase produced by *A. caespitosus* reduced kappa number only in 1.7% (xyl I), and the conditions were 10 U/g dry pulp in 2 hours (Sandrim et al., 2005).

Table 4: Ions and EDTA effect on xylanase activity produced by T. reesei QM9414 in SS medium.

	Xylanase Activity (%)	
	5 mM	**10 mM**
Cu^{2+}	106.2	78.7
Mg^{2+}	106.5	108.7
Mn^{2+}	138.8	148.7
Zn^{2+}	104.4	111.9
Fe^{3+}	89.8	93.6
Ag^{1+}	98.9	82.0
EDTA	89.9	99.2

IV. CONCLUSIONS

Sugarcane straw was evaluated as the main carbon source in axenic SmF of *T. reesei* QM 9414 to produce fibrolytic enzymes in a simple and economical bioprocess. Also, the xylanase production was successfully scaled-up from shaker flasks to bioreactor, maintaining the same culture conditions, without loss of enzyme production. This enzyme was characterized, accordingly interesting conditions for some industrial applications, mainly potential on biobleaching of kraft pulp proposes.

ACKNOWLEDGEMENTS

The authors wish to thank Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (Capes), Brazil, and Função de Amparo à Pesquisa (Fapesp - 2014/24188-1), São Paulo, Brazil, for financial support.

REFERENCES

[1] Abdullah R., Nisar K., Aslam A., Iqtedar M. & Naz S. (2015). Enhanced production of xylanase from locally isolated fungal strain using agro-industrial residues under solid-state fermentation. *Nat Prod Res. 29*, 1006-1011.

[2] Acharya S. & Chaudhary A. (2012). Bioprospecting thermophiles for cellulase production: a review. *Braz. J. of Microbiol. 43*, 844-856.

[3] Ahamed A.& Vermette P. (2008). Enhanced enzyme production from mixed cultures of *Trichoderma reesei* RUT-C30 and *Aspergillus niger* LMA grown as fed batch in a stirred tank bioreactor. *Biochem. Eng. J. 42*, 41-46.

[4] Anthony P., Harish B.S., Jampala P., Ramanujam S. & Uppuluri K.B. (2016). Statistical optimization, purification and applications of xylanase produced from mixed bacteria in a solid liquid fermentation using *Prosopis juliflora*. *Biocatal. Agric. Biotechnol. 8,* 213-220.

[5] Aquino G.S., Conti Medina C., Costa D.C., Shahab M. & Santiago A.D. (2017). Sugarcane straw management and its impact on production and development of ratoons. (Supplement C), *Ind Crops Prod.102*, 58-64.

[6] Bailey M.J., Buchert J. & Viikari L. (1993). Effect of pH on production of xylanase by *Trichoderma reesei* on xylan- and cellulose-based media. Appl Microbiol Biotechnol. 40, 224-229.

[7] Blanco A., Vidal T., Colom J.F. & Pastor F.I. (1995). Purification and properties of xylanase A from alkali-tolerant *Bacillus* sp. strain BP-23. *Appl Environ Microbiol. 61*, 4468-4470.

[8] Campioni T.S., Moreira L.J., Moretto E., Nunes N.S.S. & Oliva-Neto P. (2019). Biobleaching of Kraft pulp using fungal xylanases produced from sugarcane straw and the subsequent decrease of chlorine consumption. *Biomass and Bioenergy, 121*, 22-27.

[9] Carpio L.G.T. & Souza F.S. (2019). Competition between Second-Generation Ethanol and Bioelectricity using the

Residual Biomass of Sugarcane: Effects of Uncertainty on the Production Mix. *Molecules, 24*, 369.

[10] Carvalho A.F.A., Boscolo M., Silva R., Ferreira H. & Gomes E. (2010). Purification and characterization of the α-glucosidase produced by thermophilic fungus *Thermoascus aurantiacus* CBMAI 756. *J Microbiol. 48*, 452-459.

[11] Carvalho A.F.A., Gonçalves A.Z., Silva R. & Gomes E. (2006). A specific short dextrin-hydrolyzing extracellular glucosidase from the thermophilic fungus T*hermoascus aurantiacus* 179-5. *J Microbiol. 44*, 276-283.

[12] Carvalho A.F.A., Neto P.O., Almeida P.Z., Silva J.B., Escaramboni B. & Pastore G.M. (2015). Screening of xylanolytic *Aspergillus fumigatus* for prebiotic xylooligosaccharide production using bagasse. *Food Technol Biotechnol. 53*, 428-435.

[13] Carvalho D.M., Queiroz J.H. & Colodette J.L. (2016). Assessment of alkaline pretreatment for the production of bioethanol from eucalyptus, sugarcane bagasse and sugarcane straw. *Ind Crops Prod. 94*, 932-941.

[14] Carvalho J.L.N., Nogueirol R.C., Menandro L.M.S., et al. (2017). Agronomic and environmental implications of sugarcane straw removal: a major review. *GCB Bioenergy 9*, 1181-1195.

[15] Choi J-M., Han S-S., Kim H-S. (2015). Industrial applications of enzyme biocatalysis: Current status and future aspects. *Biotechnol Adv. 33*, 1443-1454.

[16] Cobos A. & Estrada P. (2003). Effect of polyhydroxylic cosolvents on the thermostability and activity of xylanase from *Trichoderma reesei* QM 9414. *Enzyme Microb Tech. 33*, 810-818.

[17] Conab, Companhia Nacional de Abastecimento. 2019. Acompanhamento da safra brasileira de cana-de-açúcar. *Conab Safra 2018/19 4*, 1-75.

[18] Das A., Paul T., Halder S.K., et al. (2013). Production of cellulolytic enzymes by *Aspergillus fumigatus* ABK9 in wheat bran-rice straw mixed substrate and use of cocktail enzymes for deinking of waste office paper pulp. *Bioresour Technol. 128*, 290-296.

[19] Dewan S.S, 2018. (2019, October 02).Global Markets for Enzymes in Industrial Applications. Retrieved from: https://www.bccresearch.com/marketresearch/biotechnology/global-markets-for-enzymes-in-industrial-applications.html.

[20] Dhillon G.S., Oberoi H.S., Kaur S., Bansal S. & Brar S.K. (2011). Value-addition of agricultural wastes for augmented cellulase and xylanase production through solid-state tray fermentation employing mixed-culture of fungi. *Ind Crops and Prod. 34*, 1160-1167.

[21] Duff S.J.B., Cooper D.G. & Fuller O.M. (1987). Effect of media composition and growth conditions on production of cellulase and β-glucosidase by a mixed fungal fermentation. *Enzyme Microb Technol. 9*, 47-52.

[22] Fernández-Núñez E.G., Barchi A.C., Ito S., et al. (2016). Artificial intelligence approach for high level production of amylase using *Rhizopus microsporus* var. oligosporus and different agro-industrial wastes. *J. Chem. Technol. Biot. 92*, 684-692.

[23] Ferreira-Leitão V., Perrone C.C., Rodrigues J., Franke A.P.M., Macrelli S. & Zacchi G. (2010). An approach to the utilisation of CO_2 as impregnating agent in steam pretreatment of sugar cane bagasse and leaves for ethanol production, *Biotechnol. Biofuels 7*, 2-8.

[24] Figueiredo E.B., Panosso A.R., Romão R. & La Scala N. (2010). Greenhouse gas emission associated with sugar production in southern Brazil. *Carbon Balance and Manag 5*, 3.

Gankin E, (2019, October 18). Sustainability News. Retrieved from: https://www.biofutura.com/en/materials/sugarcane.

[25] Ghose T.K. (1987). Measurement of cellulase activities. *Pure Appl Chem. 59*, 257-268.

[26] Guerra G., Casado MR-LG., Arguelles J., Sánchez M.I., Manzano A.M. & Guzman T. (2006). Cellulase production with sugarcane straw by *Trichoderma citrinoviride* on solid bed. *Sugar Tech. 8*, 30-35.

[27] Guimarães N.C.A., Sorgatto M., Peixoto-Nogueira S.C., et al. (2013). Bioprocess and biotecnology: effect of xylanase from *Aspergillus niger* and *Aspergillus flavus* on pulp biobleaching and enzyme production using agroindustrial residues as substract. *Springer Plus 2*, 380.

[28] Gutierrez-Correa M., Portal L., Moreno P., Tengerdy R.P. (1999). Mixed culture solid substrate fermentation of *Trichoderma reesei* with *Aspergillus niger* on sugar cane bagasse. *Bioresour Technol. 68*, 173-178.

[29] Haab D., Hagspiel K., Szakmary K. & Kubicek C.P. (1990). Formation of the extracellular proteases from *Trichoderma reesei* QM 9414 involved in cellulase degradation. *J Biotechnol. 16*, 187-198.

[30] Haab D., Hagspiel K., Szakmary K. & Kubicek C.P. (1990). Formation of the extracellular proteases from *Trichoderma reesei* QM 9414 involved in cellulase degradation. *J Biotechnol. 16*, 187-198.

[31] Hassuani S.J., Leal M.R.L.V. & Macedo I.C. (2005). Biomass power generation: Sugar cane bagasse and trash. In: Série Caminhos para Sustentabilidade Piracicaba: PNUD-CTC, p.19-23.

[31] He J., Yu B., Fau - Zhang K., Zhang K., Fau - Ding X., Ding X., Fau - Chen D. & Chen D. (2009). Expression of endo-1, 4-beta-xylanase from *Trichoderma reesei* in *Pichia pastoris* and functional characterization of the produced enzyme. *BMC Biotechnology 16*, 56-64.

[32] Hernández-Pérez A.F., Costa I.A.L., Silva D.D.V. et al. (2016). Biochemical conversion of sugarcane straw hemicellulosic hydrolyzate supplemented with co-substrates for xylitol production. *Bioresour Technol. 200*, 1085-1088.

[33] Isil S., Nilufer A. 2005. Investigation of factors affecting xylanase activity from *Trichoderma harzianum* 1073 D3. *Braz Arch Biol Technol. 48*, 187-193.

[34] Jänis J., Rouvinen J., Leisola M., Turunen O. & Vainiotalo P. (2001). Thermostability of endo-1,4-β-xylanase II from *Trichoderma reesei* studied by electrospray ionization Fourier-transform ion cyclotron resonance MS,

hydrogen/deuterium-exchange reactions and dynamic light scattering. *Biochem J. 356*, 453.

[35] Knob A., Beitel S.M., Fortkamp D., Terrasan C.R.F., Almeida A.F. 2013. Production, purification, and characterization of a major *Penicillium glabrum* xylanase using Brewer's spent grain as substrate. *BioMed Research International 728735*, 8.

[36] Knob A., Fortkamp D., Prolo T., Izidoro S.C., Almeida J.M. 2014. Agro-residues as alternative for xylanase production by filamentous fungi. *BioResources 9*, 5738-5773.

[37] Leal M.R.L.V., Galdos M.V., Scarpare F.V., Seabra J.E.A., Walter A., Oliveira C.O.F. (2013). Sugarcane straw availability, quality, recovery and energy use: A literature review. *Biomass and Bioenergy 53*, 11-19.

[38] Li S., Yang X., Yang S., Zhu M., Wang X. 2012. Technology prospecting on enzymes: application, marketing and engineering. *Comput. Sruct. Biotec. J. 2*, 1-11.

[39] Lopéz G. & Estrada P. (2014). Effect of temperature on xylanase II from *Trichoderma reesei* QM 9414: A calorimetric, catalytic, and conformational study. *Enzyme Res. 2014*, 16.

[40] Macedo E.P., Cerqueira C.L.O., Souza D.A.J., Bispo A.S.R., Coelho R.R.R. & Nascimento R.P. (2013). Production of cellulose-degrading enzyme on sisal and other agro-industrial residues using a new Brazilian actinobacteria strain *Streptomyces* sp. SLBA-08. *Braz J of Chem Eng. 30*, 729-735.

[41] Marín, F.R. et al. (2007). By-products from different citrus processes as a source of customized functional fibres. *Food Chem, 100*, 736-741.

[42] Miller G.L., Blum R., Glennon W.E. & Burton A.L. (1960). Measurement of carboxymethylcellulase activity. *Anal Biochem. 2*.

[43] Nagar S., Gupta V.K., Kumar D., Kumar L., Kuhad R.C. (2010). Production and optimization of cellulase-free, alkali-stable xylanase by *Bacillus pumilus* SV-85S in submerged fermentation. *J Ind Microbiol Biotechnol. 37*, 71-83.

[44] Nagar S., Mittal A., Gupta V.K. 2012. Enzymatic clarification of fruit juices (Apple, Pineapple, and Tomato) using purified *Bacillus pumilus* SV-85S xylanase. *Biotechnol Bioproc E.17*,1165-1175.

[45] Nunes L.V., Correa F.F.B., Oliva-Neto P., et al. (2017). Lactic acid production from submerged fermentation of broken rice using undefined mixed culture. *World J Microbiol Biotechnol. 33*, 79.

[46] Rahman, A. et al. (2017). Pretreatment of Wheat Bran for Suitable Reinforcement in Biocomposites. *J. Renew. Mater. 5*, n. Suppl 1, 62-73.

[47] Robl D., Delabona P.S., Costa P.S., et al. (2015). Xylanase production by endophytic *Aspergillus niger* using pentose-rich hydrothermal liquor from sugarcane bagasse. *Biocatal Biotransfor. 33*, 175-187.

[48] Salmon, D.N.X., Fendrich, R.C., Cruz, M.A., Montibeller, V.W., Vandenberghe, L.P.S., Soccol C.R. & Spier, M.R. (2016). Bioprocess for phytase production by *Ganoderma* sp. MR-56 in different types of bioreactors through submerged cultivation. *Biochem. Eng. J. 114*, 288-297.

[49] Sandrim V.C., Rizzatti A.C.S., Terenzi H.F., Jorge J.A., Milagres A.M.F. & Polizeli M.L.T.M. (2005). Purification and biochemical characterization of two xylanases produced by *Aspergillus caespitosus* and their potential for kraft pulp bleaching. *Process Biochem. 40*, 1823-1828.

[50] Sharma M., Mehta S. & Kumar A. (2013). Purification and characterization of alkaline xylanase secreted from *Paenibacillus macquariensis*. *Adv Microbiol. 3*, 32-41.

[51] Silva D.F., Camargo R., Carvalho A.F.A. & Oliva-Neto P. (2013). Cellulolytic enzyme production by the fungi *Trichoderma reesei* QM9414 and *Trichoderma reesei* CCT2768 using citrus pulp. *Poster presented at the 6th Symposium of Applied Microbiology* Rio Claro, SP, Brazil.

[52] Silva D.F., Hergesel L.M., Campioni T.S., Carvalho A.F.A. & Oliva-Neto P. (2018). Evaluation of different biological and chemical treatments in agroindustrial residues for the production of fungal glucanases and xylanases. *Process Biochem. 67*, 29-37.

[53] Silva L.A.O., Carmona E.C. [2008]. Production and characterization of cellulase-free xylanase from *Trichoderma inhamatum*. Appl Biochem Biotechnol.150,117-125.

[54] Silva, L.A.O., Terrasan C.R.F., Carmona E.C. (2015). Purification and characterization of xylanases from *Trichoderma inhamatum*. *Electron J Biotechnol.18*, 307-313.

[55] Szczerbowski, D. et al. (2014). Sugarcane biomass for biorefineries: Comparative composition of carbohydrate and non-carbohydrate components of bagasse and straw. *Carbohydr. Polym. 114*, 95-101.

[56] Terrasan C.R.F., Temer B., Sarto C., Silva Júnior F.G., Carmona E.C. (2013). Xylanase and β-xylosidase from *Penicillium janczewskii*: production, physico-chemical properties, and application of the crude extract to pulp biobleaching. *BioResources 8*, 1292-1305.

[57] Wen Z., Liao W. & Chen S. Production of cellulase/β-glucosidase by the mixed fungi culture of *Trichoderma reesei* and *Aspergillus phoenicis* on dairy manure. 2005. In: Davison B.H., Evans B.R., Finkelstein M., McMillan J.D., (Eds). Twenty-Sixth Symposium on Biotechnology for Fuels and Chemicals, Humana Press, Totowa, pp. 93-104.

[58] Xiong H., Fenel F., Leisola M., Turunen O. 2004. Engineering the thermostability of *Trichoderma reesei* endo-1,4-β-xylanase II by combination of disulphide bridges. *Extremophiles 8*, 393-400.

[59] Xiong L., Kameshwar A.K.S., Chen X., et al. (2016). The ACEII recombinant *Trichoderma reesei* QM9414 strains with enhanced xylanase production and its applications in production of xylitol from tree barks. *Microb Cell Fact.15*, 215.

https://dx.doi.org/10.22161/ijeab.51.31 *ISSN: 2456-1878*

[60] Zhang L., Wang X., Ruan Z., et al. (2014). Fungal cellulase/xylanase production and corresponding hydrolysis using pretreated corn stover as Substrates. *Appl Biochem Biotechnol*. *172*, 1045-1054.

[61] Zhou P., Zhu H., Yan Q., Katrolia P., Jiang Z. 2011. Purification and properties of a psychrotrophic *Trichoderma* sp. xylanase and its gene sequence. *Appl Biochem Biotechnol*. *164*, 944-956.

https://dx.doi.org/10.22161/ijeab.51.31 ISSN: 2456-1878

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

Utilization of Instructional Materials in Teaching Chemistry in Senior Secondary Schools in Katsina Metropolis

Yazid, Rumah Lawal*; Abdulmumin, Abdulsalam Rumah; Jamilu, Amadi

Faculty of Education, Department of Education, Uumaru Musa Yar'adua University Katsina, Katsina State, Nigeria
**Corresponding Author*
The research is financed by Asian Development Bank. No. 2006-A171(Sponsoring information)

Abstract**—** *This study investigated the Utilization of Instructional Materials in Teaching Chemistry in Senior Secondary Schools in Katsina metropolis. The study was conducted with three research objectives, three research questions and three null hypotheses. The study adopted survey research design and used questionnaire and checklist as instrument for data collection. The sample for the study was arrived at using random sampling technique, hence the total of twenty four (24) chemistry teachers and three hundred and seventy (370) students were used as research sample. The analysis of the data collected was done using both descriptive and inferential statistics. Findings among others shows that: there is no significant difference in the availability of instructional materials for teaching chemistry in senior secondary schools in Katsina metropolis as the observed p value 0.310 is greater than the alpha value (p=0.310>0.05), and that there is a significant difference in the utilization of instructional materials in teaching chemistry in senior secondary schools in Katsina metropolis as the observed p value 0.027 is less than the alpha value (p=0.027<0.05). It is recommended that adequate instructional materials for teaching of chemistry in senior secondary schools in Katsina metropolis should be provided by the ministry of education and that teachers should improvise where instructional materials are not available to teach chemistry in the senior secondary schools in Katsina metropolis, among others.*

Keywords**— China insurance industry, Foreign fund, Challenge.**

I. INTRODUCTION

Instructional materials are objects or devices that help the teacher to make learning meaningful to the learners Ikerionwu (2000). The importance of instructional materials in enhancing effective teaching and learning of science, chemistry in particular cannot be undermined due to positive impact it has on students' performance. Instructional materials assist the teacher to achieve the stated goals and objectives. Instructional materials of all kind appeal to the sense organs during teaching and learning Agina-Obu (2005). Instructional materials which are educational inputs are of vital importance to teaching of any subject in the school curriculum.

In his own perspective, Abdu-Raheem (2014) acknowledged that instructional materials are used by teachers to aid explanations and make learning of subject matter understandable to students during teaching and learning process. Instructional materials are essential and significant tools need for teaching and learning to promote teacher's efficiency and capture the student's attention in classroom situation. Kochhar (2012) supported that instructional materials are very significant learning and teaching tools. He suggested there is need for teachers to find necessary materials for instruction to supplement what textbook provide in order to broaden concepts and arouse student's interests in the subject. Fadeiye (2005) saw instructional materials as visual and audio-visual aids, concrete or non concrete, used by teachers to improve the quality of teaching and learning activities.

However, Akinleye (2010) attested that effective teaching and learning requires a teacher to teach the student with instructional materials and use practical activities to make learning more vivid, logical, realistic and pragmatic. Despite the fact that instructional materials are essential tools that can make learning, practical and knowledge acquisition easier they are not readily available in Nigerian

secondary schools leading to low level of performance of learners in government examinations. This prompted the researcher to embark on this research whose focus is to survey the utilization of instructional materials in teaching chemistry in srnior secondary schools in Katsina metropolis.

Instructional materials make teaching and learning more meaningful, understandable and easy. But in spite of the benefits of instructional materials to teaching and learning, the scarcity and inadequate utilization of the instructional materials has hindered, to some extent, the efficiency of teaching and learning of chemistry. Instructional materials make learning meaningful and help to improve students' academic achievement. However these advantages of instructional materials have not reflected in the education system because of the dearth of these instructional materials in our schools. Most of the teachers do not even care to use instructional materials, they only depend on the old traditional method. This problems lead to students' massive failure in chemistry examinations, especially the school leaving certificate examination. Also Nov/Dec result of 2014 indicates that 51.62 percent out of 28,250 candidates who sat for chemistry passed with credit, indicating only half of the candidates passed with credit in chemistry that year.

It is in view of the above discussion, the researcher intended to embark on a survey research whose attention focuses on utilization of instructional materials in teaching chemistry in senior secondary schools in Katsina metropolis.

II. THE OBJECTIVES OF THIS STUDY

1. Find out the availability of instructional materials in teaching chemistry in senior secondary schools in Katsina metropolis.
2. Find out the extent of utilization of instructional materials in teaching chemistry in senior secondary schools in Katsina metropolis.
3. Find out the difference in the utilization of instructional materials between male and female teachers in teaching chemistry in senior secondary schools in Katsina metropolis.

This research project is expected to contribute positively in the areas of knowledge expansion, utilization of instructional materials towards effective teaching and learning chemistry. This would be significant to all stake holders in education such as, educational administrators, teachers, students, ministry of education and the society at large in identifying educational problems and challenges, there by coming up with possible solution and strategies to these problems.

III. RESEARCH METHODOLOGY

The research design adopted for this study is descriptive survey method. The survey method is adopted because the research involved collecting data from teachers and students hence, make generalization. The design is considered appropriate because it is thought to be such a design that will enable the researcher in identifying the characteristics of the phenomena under study through administration of questionnaires and working by checklist of instructional materials. Survey method is a method characterized by the selection of random sample from a large and small population in order to obtain empirical knowledge of contemporary nature.

3.1 Research Population

The population for this study stands at 10,520 students offering chemistry in public senior secondary schools in Katsina metropolis. And 24 Chemistry teachers in public senior secondary schools in Katsina metropolis. Thus the population of the study with respect to chemistry students is summarized in the table below;

Table 1. List of Schools and Their students' Population.

S/N	Schools	SS1	SS2	SS3	Total
1.	Dikko College Katsina	181	205	161	547
2.	Family Support Secondary School Katsina	99	72	102	273
3.	Government College Katsina, Day Wing	401	465	281	1147
4.	Government Girls College Senior Katsina	290	229	232	751

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

5.	Government Secondary School Dutsin-Safe	139	118	62	319
6.	Government Senior Secondary School K/Kaura	263	365	388	1016
7.	Government Senior Secondary School K/Yandaka	690	401	264	1355
8.	Government School For the Blind Katsina	97	95	97	608
9.	Government Pilot Senior Secondary School K/Sauri	719	712	621	2052
10.	Katsina College Katsina Senior	410	441	489	1340
11.	Sir Emeka Offer Senior Sec. School Kambarawa	330	281	239	850
12.	Sir Usman Nagogo College of Arabic and Islamic Studies	242	182	163	587
	Total	**3855**	**3566**	**3099**	**10520**

(Source: Zonal Education Quality Assurance Office, Katsina Zone 2017)

Table 2. Population of Chemistry teachers with respect to school

S/N	**Schools**	**Number of teachers**
1.	Dikko College Katsina	2
2.	Family Support Sec. School Katsina	1
3.	Government College Katsina Day Wing	4
4.	Government Girls College Senior Katsina	4
5.	Government Secondary School Dutsin-Safe	–
6.	Government Senior Secondary School K/Kaura	2
7.	Government Senior Secondary School K/Yandaka	1
8.	Government School For the Blind Katsina	2
9.	Government Pilot Senior Secondary School K/Sauri	1
10.	Katsina College Katsina Senior	2

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

11. Sir Emeka Offer Senior Sec. School Kambarawa	4
12. Sir Usman Nagogo College of Arabic And Islamic Studies	1
Total	**24**

(Source: Zonal Education Quality Assurance Office, Katsina Zone 2017)

3.2 Sample and Sampling Techniques

To determine sample size appropriate for this study, the researcher first consulted Research Advisers (2006) table of sample, keeping in mind the total population size of the students (10,520). The table suggests that a population size of 10,000 should have sample size of 370. The table also equally suggest that for a population size of (24), the sample size shall be (20). Random sampling technique was used in selecting the schools.

Table 3. Sampled Schools

S/N	School	Students' population	Number of sampled students
1.	GSSS K/Yandaka	1355	169
2.	GSSS K/Kaura	1016	127
3.	Sir Usman Nagogo College of Arabic and Islamic Studies	587	74
Total		**2,958**	**370**

However, in the case of teacher-respondents; looking at the number of chemistry teachers (24) in senior secondary schools in Katsina metropolis, the researcher decided to include all the chemistry teachers found in the sampled schools.

3.3 Research Instruments

The instrument used for this study was questionnaire and checklist constructed by the researcher which was subjected to validity. The researcher used two sets of questionnaire; one set for students, and one for teachers. Both the teachers' questionnaires and students' questionnaires contained fifteen (15) items each; and are designed on a five-point Likert scale (comprising of

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

Strongly Agree, Agree, Undecided, Disagree and Strongly Disagree) in order to supply data as regards teachers and students opinions on the utilization of instructional materials in teaching chemistry in senior secondary schools in Katsina metropolis.

The (25) item checklist contained some instructional materials and equipment in teaching chemistry. The checklists are intended to assist the researcher in appropriately taking the inventory of and cross checking the available instructional materials and equipments in senior secondary schools in Katsina metropolis.

3.4 Validity of the Instruments

A measuring instrument is considered valid only when it measures truly and accurately what it intends to measure. In this case, content and face validity of the instrument was determined by lecturers in the Department of Education of Umaru Musa Yar'adua University for validation. Meanwhile, all corrections and modifications suggested by the validators are to be effected.

3.5 Pilot Testing for Reliability of the Instruments

Further to the validation of the instrument, the researcher conducted a pilot study on 50 senior secondary students of Albarka Secondary Schools (which is not part of the population of the study and not among the sample) and computed split-half reliability (reliability of internal consistency). To do so the researcher systematically split the scores obtained into two equal halves using odd and even number position of the items. The corresponding two set of scores obtained were then correlated using Pearson product moment correlation method. The r value obtained was

0.52 which indicates the reliability of the half test, the **r** value was then substituted into the spearman's brown formula to obtain the reliability of the full test. The **R** value obtain was 0.68 indicating strong reliability of the instrument.

3.6 Procedure for Data Collection

The teachers' questionnaires and students' questionnaires were self-administered by the researcher in all the three sampled schools. Again, the checklists was also used by the researcher in taking down the availability of equipment and materials in the sampled schools.

The researcher also asked for the assistance of teachers to assist in administering the questionnaires to the students. In order to administer the instrument appropriately, systemically sampling technique was employed by the used of the class register, the researcher sampled out students by which their number ends with even number. Explanation was equally done by the researcher on how to respond to the questionnaire before students were asked to respond to the items on the questionnaire. Teacher's questionnaire was administer to them in the staff rooms. The researcher instantly collected back the scripts upon the completion of filling the instruments.

3.7 Procedure for Data Analysis

The data to be obtained are merely responds which will be converted to numbers, which may be meaningless if not reduced to usable and meaningful form. In terms of answering research questions, the researcher decided to employ descriptive statistics. While for testing hypotheses Ho1 and Ho2 analysis of variance ANOVA would be employ, while for Ho3 t-test independent sample would be employ for determining significant differences between two set of variables or phenomena. This is because the task involves the analysis of data from fairly large sample size of respondents. More also, the data collected is a nominal data and a continuous data and not a discrete one, in which case a non-parametric tool is the best.

IV. RESULTS & DISCUSSIONS

4.1 Presentation of Data

The data collected for this research is presented and analyzed using statistical package for social science SPSS in order to answer the research question and test the hypothesis with a view of knowing the utilization of instructional materials in teaching chemistry in senior secondary schools in Katsina metropolis, thus the data collected is presented in the following ways:

4.2 Data Analysis

Table 4. showing the availability of Instructional materials in SUNCAIS

S/N	Name of equipment/material	Absent	Present Confirmed	Number
1.	Teachers' preparatory office	Yes	1	

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

2.	Charts			Yes	3
3.	Conical flasks			Yes	30
4.	Tripod and retort stand			Yes	20
5.	Test tubes			Yes	30
6.	Watch glasses	Yes			
7.	Bunsen burner		Yes		
8.	Funnels			Yes	20
9.	Graduated cylinders			Yes	10
10.	Volumetric flasks			Yes	20
11.	Droppers			Yes	10
12.	$25cm^3$ pipette			Yes	25
13.	Burette			Yes	23
14.	Ring stands, rings, and clamps			Yes	13
15.	Tongs and forceps			Yes	11
16.	Spatulas			Yes	7
17.	Weighing balance			Yes	3
18.	Petridish			Yes	17
19.	Reagent bottles			Yes	21
20.	Test tube rack			Yes	7
21.	Beaker			Yes	27
22.	Thermometer			Yes	4
23.	Blue litmus paper			Yes	4 packs
24.	Red litmus paper			Yes	4 packs
25.	Filter paper			Yes	5 packs

From the above data collected from the check list it can be seen that all the 25 instructional materials are available in SUNCAIS with the exception of watch glasses and Bunsen burners.

Table 5. showing the availability of instructional materials in GSSS K/KAURA

S/N	**Name of equipment/material**	**Absent**		**Present**	
Confirmed		**Number**			
1.	Teachers' preparatory office			Yes	2
2.	Charts			Yes	2
3.	Conical flasks			Yes	30
4.	Tripod and retort stand			Yes	30
5.	Test tubes			Yes	20
6.	Watch glasses			Yes	10
7.	Bunsen burner			Yes	15
8.	Funnels			Yes	16

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

9.	Graduated cylinders	Yes		
10.	Volumetric flasks		Yes	15
11.	Droppers		Yes	5
12.	$25cm^3$ pipette		Yes	20
13.	Burette		Yes	20
14.	Ring stands, rings, and clamps		Yes	20
15.	Tongs and forceps		Yes	5
16.	Spatulas		Yes	12
17.	Weighing balance		Yes	1
18.	Petridish		Yes	10
19.	Reagent bottles		Yes	20
20.	Test tube rack		Yes	4
21.	Beaker		Yes	20
22.	Thermometer		Yes	11
23.	Blue litmus paper		Yes	4 packs
24.	Red litmus paper		Yes	4 packs
25.	Filter paper		Yes	5 packs

From the above data collected from the check list it can be seen that all the 25 instructional materials are available in G.S.S.S K/Kaura. With the exception of graduated cylinders.

Table 6. showing the availability of instructional materials in GSSS K/YANDAKA

S/N	**Name of equipment/material**	**Absent**	**Present**
Confirmed	**Number**		
1.	Teachers' preparatory office	Yes	2
2.	Charts	Yes	15
3.	Conical flasks	Yes	30
4.	Tripod and retort stand	Yes	15
5.	Test tubes	Yes	10
6.	Watch glasses	Yes	3
7.	Bunsen burner	Yes	1
8.	Funnels	Yes	20
9.	Graduated cylinders	Yes	5
10.	Volumetric flasks	Yes	3
11.	Droppers	Yes	10
12.	$25cm^3$ pipette	Yes	12
13.	Burette	Yes	20
14.	Ring stands, rings, and clamps	Yes	13
15.	Tongs and forceps	Yes	2
16.	Spatulas	Yes	10

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

17.	Weighing balance	Yes	1
18.	Petridish	Yes	5
19.	Reagent bottles	Yes	20
20.	Test tube rack	Yes	5
21.	Beaker	Yes	28
22.	Thermometer	Yes	8
23.	Blue litmus paper	Yes	5 packs
24.	Red litmus paper	Yes	7 packs
25.	Filter paper	Yes	4 packs

From the above data collected from the check list it can be seen that all the 25 instructional materials are available in G.S.S.S K/Yandaka.

To adequately answer the research question a table of percentage on the availability of instruction materials in the 3 schools was computed as follows;

Table 7. showing the percentage of availability of instructional materials.

S/N	Schools	% Available	% Unavailable
1.	SUNCAIS	92%	8%
2.	GSSS K/Kaura	96%	4%
3.	GSSS K/Yandaka	100%	0%
	TOTAL	96%	4%

From table 7, above, it is clear that the percentage of the available equipment and material in the three schools is 96%, On the other hand, the percent of the unavailable equipment and materials in the three schools is 4%.
Meanwhile to answer this research question; all the 25 listed instructional materials are adequately available in the senior secondary schools of Kasina Metropolis with the exception Bunsen burner, graduated cylinder and watch glasses.

Table 8. Responses of students on utilization of instructional materials in teaching chemistry

S/N	ITEM	SA	A	U	DA	SD
1.	Our chemistry laboratory is adequately utilized in teaching chemistry.	13.24%	2.97%	2.97%	21.1%	64.28%
2.	Our chemistry teacher use Periodical	28.11%	4.95%	0%	10.81%	56.49%

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

	charts during chemistry lesson.					
3.	Our chemistry teacher use Graphical materials (map and charts) during chemistry lesson.	7.03%	1.89%	1.08%	59.2%	30.81%
4.	Volumetric flask is adequately utilized In teaching chemistry	13.24%	3.51%	2.97%	31.89%	48.38%
5.	Weighing balance is adequately utilized in teaching chemistry.	2.97%	0.81%	1.08%	14.04%	81.08%
6.	Conical flasks are adequately utilized in teaching chemistry.	2.16%	5.4%	0%	16.21%	76.22%
7.	Test tubes and racks are adequately utilized in teaching chemistry.	10.81%	4.59%	0%	56.49%	28.11%
8.	Burettes are adequately utilized in teaching chemistry.	0.54%	5.95%	1.35%	13.51%	78.64%
9.	Pipettes are adequately utilized in teaching chemistry.	0%	0.27%	0%	4.3%	95.4%
10.	Measuring cylinders are adequately utilized in teaching chemistry.	6.48%	2.97%	0.54%	72.97%	17.08%
11.	Heat sources such as Bunsen burner ınd portable burner are adequately utilized in our chemistry laboratory.	1.08%	0.27%	0.54%	2.97%	95.14%
12.	Beakers are adequately utilized in our Chemistry teachers improvise instructional materials and equipment during chemistry lessons.	0% 0.54%	0% 0.27%	0% 0%	2.16% 1.08%	97.84% 98.11%
	AVERAGE	**6.63%**	**2.61%**	**0.81%**	**24.42%**	**64.89%**

From the tables above, it is clear that majority of the students (64.89%) strongly disagreed and

24.42% disagreed with the fact that chemistry teachers adequately utilize instructional materials in teaching chemistry in the schools under study.

Table 9. Responses of Teachers on utilization of instructional materials in teaching chemistry

S/N	*ITEM*	*SA*	*A*	*U*	*DA*	*SD*
		%	**%**	**%**	**%**	**%**
1.	Laboratory is adequately utilized in teaching chemistry.	14.3%	0%	0%	21.1%	64.28%
2.	Periodical charts are adequately utilized in teaching chemistry.	35.7%	42.9%	14.3%	7.1%	0%

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

3.	Graphical materials (map and charts) are adequately utilized in teaching chemistry.	14.3%	21.1%	7.1%	28.6%	28.6%
4.	Volumetric flask is adequately utilized in teaching chemistry	42.9%	0%	0%	7.1%	50%
5.	Weighing balance is adequately utilized in teaching chemistry.	14.3%	7.1%	0%	35.7%	42.9%
6.	Conical flasks are adequately utilized in teaching chemistry.	64.28%	21.1%	0%	0%	14.3%
7.	Test tubes and racks are adequately utilized in teaching chemistry.	50%	42.9%	7.1%	0%	0%
8.	Burettes are adequately utilized in teaching chemistry.	0%	0%	0%	42.9%	57.1%
9.	Pipettes are adequately utilized in teaching chemistry.	78.6%	0%	0%	7.1%	14.3%
10.	Measuring cylinders are adequately utilized in teaching chemistry.	0%	0%	7.1%	7.1%	85.71%
11.	Heat sources such as Bunsen burner and portable burner are adequately utilized in our chemistry laboratory.	14.2%	7.1%	0%	21.1%	87.1%
12.	Beakers are adequately utilized in our chemistry laboratory.	7.1%	0%	7.1%	35.7%	50%
13.	Chemistry teachers improvise instructional materials and equipment during chemistry lessons.	0%	42.9%	57.1%	0%	0%
AVERAGE		**29.12%**	**15.33%**	**3.28%**	**16.42%**	**35.71%**

From the above table 35.71% of the teachers strongly disagreed and 29.12% strongly agreed with the fact that chemistry teachers utilize instructional materials in teaching chemistry in the schools under study.Meanwhile, the answer to this question is that 'chemistry teachers do not adequately utilize chemistry instructional materials in teaching chemistry in senior secondary schools in Katsina Metropolis

Table 10. Showing responses of both teachers and students on item 13 of the questionnaire

RESPONSE	**SA**	**A**	**U**	**DA**	**SD**
FREQUENCY	323	2	5	50	0
PERCENTAGE	84.1%	0.52%	1.30%	13.02%	0%

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

From the table above 84.1 % of the respondents strongly agreed and 13.02% of the respondent disagree with the statement "male chemistry teachers use instructional materials more than the female chemistry teachers". Therefore to answer the research question, Male chemistry teachers utilizes instructional materials in teaching chemistry more than the female chemistry teachers in senior secondary schools of Kastina Metropolis.

This research was tested using inferential statistics (one way ANOVA) with the aid of statistical package for social science (SPSS).

In the first place, data collected using five-point linkert scale such as Strongly agree, SA, Agree A, Undecided, U, Disagree, DA and Strongly disagree, SD; or extremely satisfied, satisfied, neutral, dissatisfied and extremely dissatisfied; is a nominal data. Therefore, for the purpose of this analysis codes are usually given as follows

Strongly agree - 5
Agree - 4
Undecided - 3
Disagree - 2
Strongly disagree - 1

Table 11. One Way ANOVA analyses of the mean score SUNCAIS, GSSS K/Kaura and GSSS K/Yandaka on the availability of instructional materials in teaching chemistry

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

Source	Sum of squared	DF	Mean square	Critical value	F	Sig
Between Group	188.880	2	94.440	3.0	1.189	0.310
Within Group	5719.120	72	79.432			
Total	5908.00	74				

*significant at p≤ 0.05

The result in table above shows that the schools does not differ significantly in the availability of instructional materials. The observed F value (1.189) in less than the 3.00 for the critical value at 2, 72 degree of freedom. The observed p value 0.310 is greater than 0.05 this means that the null hypotheses that there is no significant difference in the availability of chemistry instructional materials in senior secondary schools of Katsina Metropolis is accepted.

Table 12. One Way ANOVA analyses of the mean score SUNCAIS, GSSS K/Kaura and **GSSS K/Yandaka on the utilization of instructional material in teaching chemistry**

Source	Sum of square	DF	Mean square	Critical F-value	Calculated F-value	Sig
Between Group	133.053	2	66.527	3.0	1.985	0.027
Within Group	12301.220	367	33.518			
Total	12434.273	369				

*significant at p≤ 0.05

The result in table above shows that the schools differ significantly in the utilization of Instructional materials. The observed F value (1.985) in less than the 3.00 for the critical value at

2, 367 degree of freedom. The observed p value 0.027 is less than 0.05 this means that the null hypotheses that there is no significant difference in the utilization of instructional materials in teaching chemistry senior secondary schools of Katsina Metropolis is rejected.

Table 13. Independent sample t-test on the Teachers score in the utilization of instructional materials between male and

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

female teachers in teaching chemistry

Pair	N	Mean	Mean Difference	Standard deviation	T	Degree of	P
Male teachers	7	3.57	0.429	1.618	0.612	12	0.552
Female teachers	7	3.14		0.900			

*significant at p≤ 0.05

From the above table it is clear that the P value (0.552) is greater than 0.05 which signifies there is no significant difference in the utilization of instructional materials in teaching chemistry between male and female chemistry teachers therefore the null hypothesis is accepted.

4.3 Summary Major Findings

The following findings emerged from the study that;

1) There is no significant difference in the availability of instructional materials for teaching chemistry in senior secondary schools in Katsina Metropolis due to the fact that the observed p value 0.310 is greater than 0.05.
2) There is significant difference in the utilization of instructional materials for teaching chemistry in senior secondary schools in Katsina Metropolis as the observed p value 0.027 is less than 0.05.3) There is no significant difference in the utilization of instructional materials between male and female teachers of chemistry in senior secondary schools in katsina metropolis as P value (0.552) is greater than 0.05

4.4 Discussion of Result

Table 4.1, 4.2 and 4.3 shows that almost all the 25 listed instructional materials are adequately available with the exception of Bunsen burner, watch glasses and graduated cylinders, also from table 4.4 it is clear that the percentage of the available equipment and material in the three schools is 96%, On the other hand, the percent of the unavailable equipment and materials in the three schools is 4%. This indicate that there are instructional materials available for teaching chemistry in senior secondary schools in Katsina Metropolis, This implies that probability of the students of Katsina metropolis performing better is high. From empirical evidence, Edet (2008) in his study the result showed that students taught using laboratory facilities frequently achieved higher than those taught without utilizing laboratory facilities during Biology lessons.

Table 4.5 and Table 4.6 shows that teachers of chemistry do not utilize instructional materials, as majority of the students (64.89%) strongly disagreed and 24.42%

https://dx.doi.org/10.22161/ijeab.51.32 *ISSN: 2456-1878*

disagreed with the fact that chemistry teachers adequately utilize instructional materials in teaching chemistry in the schools under study while on the hand 35.71% of the teachers strongly disagreed and 29.12% strongly agreed with the fact that chemistry teachers utilize instructional materials in teaching chemistry in the schools under study. This indicate that despite the availability of instructional materials, teachers of chemistry do not adequately utilized them. This is in line with Uyagu (2009) which revealed that students performed better when appropriate and improvised materials were made available and utilized in teaching science.

Table 4.7 shows that male chemistry teachers utilizes instructional materials in teaching chemistry more than the female chemistry teachers in senior secondary schools of Kastina Metropolis. Due to the fact 84.1 % of the respondents strongly agreed and 13.02% of the respondent disagree with the statement "male chemistry teachers use instructional materials more than the female chemistry teachers".

Table 4.8 shows schools does not differ significantly in the availability of instructional materials for teaching chemistry in senior secondary school in Katsina metropolis, since observed p value 0.310 is greater than 0.05. while Table 4.9 shows that the schools differs significantly in the utilization of instructional for teaching chemistry in senior secondary schools in Katsina metropolis since the observed p value 0.027 is less than 0.05 . Thus implies that despite the availability of the instructional materials chemistry teachers do not adequately utilize them. This finding is in line with that of Opara (2008) which revealed that laboratory facilities were not utilized during chemistry teaching and learning.

Table 4.10 shows that there is no significant difference in the utilization of instructional materials between male and female teachers of chemistry in senior secondary school in Katsina Metropolis, from the table it is clear that the P value (0.552) is greater than 0.05.

V. CONCLUSION AND RECOMMENDATION

5.1 Conclusion

From the findings of the study, it can be concluded that utilization of instructional materials have influence in teaching chemistry in senior secondary schools in Katsina metropolis and that there are instructional materials available for teaching chemistry. Also, based on the finding, it can be inferred that chemistry teachers do not use instructional materials in teaching chemistry in senior secondary schools in Katsina metropolis.

5.2 Recommendations

Based on the findings of this research, the following recommendations were made:

1. Adequate instructional materials for the teaching of chemistry in senior secondary schools in Katsina metropolis should be provided by the ministry of education and school management.
2. Teachers should improvise where instructional materials are not available to teach chemistry in the senior secondary schools in Katsina metropolis.
3. Utilization of instructional materials should be encourage at all levels of education.
4. Teacher education programme should integrate materials development whereby teachers learn how to design and construct various materials and equipment which could be used for teaching- learning process.
5. Resource centres should be created in each Education zone where teachers can go to borrow teaching materials or take their students there to use the materials.

REFERENCES

[1] Abdullahi, T. (2004). *Teaching facilities and students' performance in chemistry. A case study of Secondary school students in Zaria* M.Ed thesis, A.B.U, Zaria.

[2] Abdu-Raheem, B. O. (2014). Improvisation of instructional materials for teaching and learning In secondary schools as predictor of high academic standard. *Nigerian Journal of SocialStudies, vol(17).No*(1).

[3] Abdu-Raheem, B. O. (2016). Effects of instructional materials on Secondary Schools students'academic achievement in Social Studies in Ekiti State. *World Journal of Education, vol (6).No (1).*

[4] Abiodun-Oyebanji, O., & Adu, E.T. (2007). Principals' performance in instructional supervision In the use of resources in senior secondary schools in Ekiti State. *Journal of Educational Focus,*Vol. (*1)No.*(1).

[5] Agina-Obu, T.N. (2005). The relevance of instructional materials in teaching and learning. Ajayi, Y.A. (2000). *A guide for effective teaching*. Lagos: IDEMAC Nigeria Ltd.

[6] Akinsola, M. K and Igwe, I. O. (2002). *The relative effects of Meta cognitive strategy of framing On students' achievement*

in selected difficult chemistry concept, Journal of Science Teachers Association of Nigeria, 37, (1 & 3), 20 – 28.

[7] Amaefule, A. A. (2001). Chemistry Education as a basis for Technological Development: *Gombe Technical Education Journal, 3, 63-75.*

[8] The ASSURE Model: Creating the Learning Experience." *The ASSURE Model: Creating The Learning Experience*. Pearson,n.d. Retrieved 09 july 2017 http://wps.prenhall.com/chet_smaldino_instruct_9/67/17197/4402563.cw/index.html

[9] Edet, U. B. (2008). Effect of environmental resources on students' academic performance in

[10] Biology. 49th annual conference of Science Teachers' Association of Nigeria.

[11] Esu, A.E.O., Enukoha, O.I.T., & Umorem, G. U. (2004). *Curriculum development in Nigeria for Colleges and universities*. Owerri: Whyte and Whyte Publishers.

[12] Fadeiye, J.O. (2005). *A social studies textbook for colleges and universities*. Ibadan: Akin Johnson Press and Publishers.

[13] Federal Republic of Nigeria (2004). *National Policy on Education.* Lagos: Heinemann Educational books Ltd.

[14] George, G. O. (2008). Causes of Poor Achievement in WAEC Biology Examinations in Taraba State Secondary Schools. *International Journal of Life Sciences and Technology* 21 (3).

[15] Ibrahim, S.A. (2000). Instructional Materials. *Journal–Teachers Periscope of NUT, Kogi State Wing*, Vol. 1. (2), 27-29.

[16] Ikerionwu, J.C. (2000). Importance of aids and resources in classroom teaching. Knirk & Gustafson Instructional Design model. (n.d.). Retrieved July 18, 2017, from http://lts.ncsu.edu/guides/instructional_design/selecting_models2.htm

[17] Kochhar, S. K. (2012). *The teaching of social studies.* New Delhi, India. Sterling Publishers Private Limited.

[18] Maduekwe, A. N. (2006). Perspective on Teacher Education and ICTs in the Language Classroom, V. B. (ed). *An Introduction to Educational Technologies in Education* Lagos: Sibon books Ltd.

[19] Mba, T. N. (2004): *Adult education and development.* Port Harcourt: Pearl Publisher. Mberekpe Augustine, Chukwunyeremunwa. (2013) Effect of students improvised instructional Materials on senior secondary school students' achievement in biology. University of Nigeria Nsukka.

[20] NERDC, (2009). Workshop on Difficult Concepts Biology Group report. Nigeria Educational Research and Development Council Lagos.

[21] Nwagbo, C.R. (2006). Effects of two teaching methods on achievement and Attitude to Biology Of Students of different levels of scientific literacy. *International Journal of Educational Research.* Retrieved from www.elsevier.com/locate/ijdures. On 13th April 2017.

[22] Nzewi,U. M, and Nwosu, A. A. (2009). *Course Guide, EDU 236: Biology Methods*. National Open University of Nigeria.

[23] Ojokuku, O.G. (2012). *Understanding Chemistry,* Zaria:MacChin multimedia Designers, Press On Chem resources.

[24] Ojokuku, O.G. (2001). *Practical Chemistry, for schools and colleges.* Ibadan: Gbabeks Publishers Ltd.

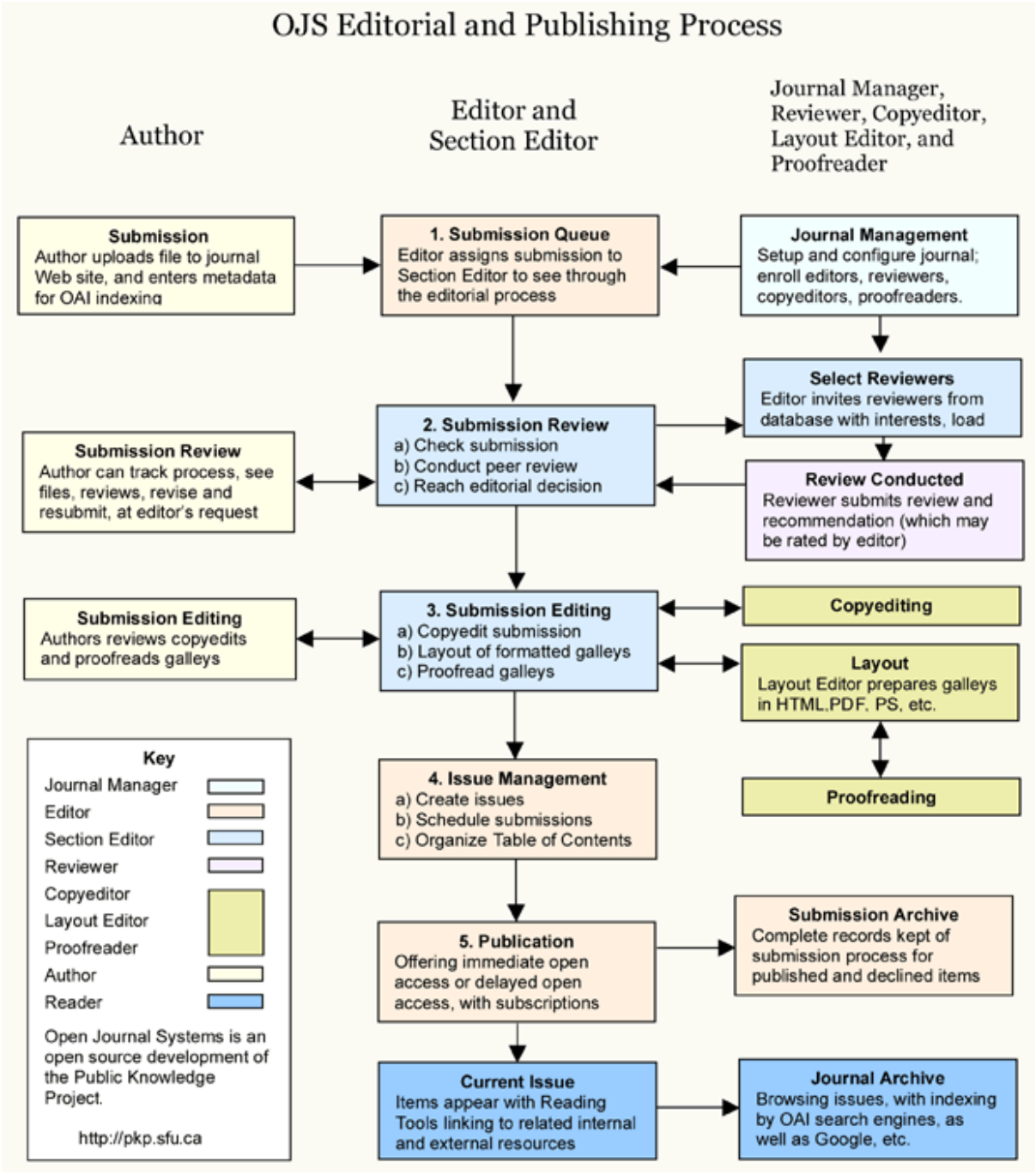
[25] Oladipo, F.O. (2001). *Introduction to Agricultural Journalism and audio-visual methods.* Ilorin: KWCOED, Ilorin.

[26] Olumorin, C. O., Yusuf, A., Ajidagba, U. A., & Jekayinfa, A. A. (2010). Development of Instructional materials from local resources for art-based courses. *Asian Journal of Information Technology, Vol. (9) No (.2),* http://dx.doi.org/10.3923/ajit.2010.107.110.

[27] Oluwagbohunmi, M.F., & Abdu-Raheem, B.O. (2014). Sandwich undergraduates' problem of Improvisation of instructional materials in social studies: The case of Ekiti State University

[28] Opara, M. F. (2008). Utilization of laboratory facilities and students' academic performance Unpublished M.Ed. thesis of Anambra State University, Nigeria.

[29] Uyagu, B.G. (2009) Effects of Instructional Materials Usage and Teachers' Quality on Students Performance in Science in Senior Secondary Schools in Zaria LGA of Kaduna State. Unpublished M.Ed. Thesis, A.B.U. Zaria – Nigeria.



~OJS Workflow~

Important links:

Paper Submission Link:
OJS:
https://ijeab.com/ojs/index.php/ijeab/about/submissions
https://ijeab.com/submit-paper/
Editorial Team:
https://ijeab.com/editorial-board/

Peer Review Process:
https://ijeab.com/peer-review-process/
Publication Ethics:
https://ijeab.com/publication-policies-and-ethics/
Author Guidelines:
https://ijeab.com/author-guidelines/
Join Us a Reviewer:
https://ijeab.com/join-us/

Journal Indexed and Abstracted in:

- Qualis-CAPES -Brazil
- Normatiza (Under Review)
- Bielefeld Academic Search Engine(BASE)
- Aalborg University Library (Denmark)
- WorldCat: The World's Largest Library Catalog
- Semantic Scholar
- J-Gate
- Open J-Gate
- CORE-The world’s largest collection of open access research papers
- JURN
- Microsoft Academic Search
- Google Scholar
- Kopernio - powered by Web of Science
- Pol-Index
- PBN(Polish Scholarly Bibliography)Nauka Polaska
- Scilit, MDPI AG (Basel, Switzerland)
- Tyndale University College & Seminary
- indiana Library WorldCat
- CrossRef DOI-10.22161/ijeab
- Neliti - Indonesia's Research Repository
- Journal TOC
- Dimensions.ai: Re-imagining discovery and access to research
- Citeseerx
- Massachusetts Institute of Technology (USA)
- Simpson University (USA)
- University of Louisville (USA)
- Biola University (USA)
- IE Library (Spain)
- Mount Saint Vincent University Library (Halifax, Nova Scotia Canada)
- University Of Arizona (USA)
- INDIANA UNIVERSITY-PURDUE UNIVERSITY INDIANAPOLIS (USA)
- Roderic Bowen Library and Archives (United Kingdom)
- University Library of Skövde (Sweden)
- Indiana University East (campuslibrary (USA))
- Tilburg University (The Netherlands)
- Williams College (USA)
- University of Connecticut (USA)
- Brandeis University (USA)
- Tufts University (USA)
- Boston University (USA)
- McGill University (Canada)
- Northeastern University (USA)
- BibSonomy-The blue social bookmark and publication sharing system
- Slide Share
- Academia
- Archive
- Scribd
- SJIF-InnoSpace
- ISSUU
- Research Bib
- DRJI
- journal-repository

Platform & workflow by OJS / PKP

Infogain Publication
International Journal of English, Literature and Social Science (IJELS)
83/197, Sector-8, Pratap Nagar, Jaipur, India