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FOREWORD

I am pleased to put into the hands of readers Volume-7; Issue-5: September-October 2022 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
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Micronutrient Concentrations of Cassava Continuously Cultivated Soils in Ezinihitte Mbaise LGA Imo State, Nigeria

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Abstract— *The study was conducted to determine the concentrations of micronutrients (Copper (Cu), Zinc (Zn), Iron (Fe), Molybdenum (Mo) and Manganese (Mn)) in soils of Ezinihitte Mbaise LGA Imo State. The research was conducted in cassava continuously cultivated areas Obizi, Eziudo, Onisha and Udo in Ezinihitte to trace the role of micronutrient in the decline of Cassava yield in the area. Samples were collected randomly using soil auger from each locations at a depth of 0-30 centimeter (cm). Control (reference) soils were collected the same way but from fallow land of 5years old. Samples were treated and analyzed routinely and micronutrients (Cu, Zn, Fe, Mo and Mn) determined using Atomic Adsorption Spectrophotometer (AAS). The result (table 1) revealed that Samples have same soil texture -Loamy Sand (LS) but differ in pH significantly (p<0.05). The samples are acid soils of moderately acidity, and levels ranges from 5.00 to 5.44 (lowest to highest) with mean value of 5.21. For % Nitrogen N, % Organic Matter (OM), % Base Saturation (BS), Available Phosphorus (Av. P), Potassium (K), Magnesium (Mg), Calcium (Ca), Sodium (Na), EA (Ea) and Effective Cation Exchange capacity (ECEC). They were significantly affected by cultivation and have mean values (P -1.57, N -1.33, K-1.60, OM 2.79, BS-74.3, Mg-2.46, Ca-1.47, K-0.15, Na-0.13, Ea-1.48 and ECEC-5.47) (table 1). This is a common characteristics of moderate to low fertility soils. The micronutrient levels were also significantly different. Cassava cultivation affected micronutrients levels significantly at p<0.05 (table2). The decrease were observed in all samples, and were significantly lower when compared with control. For Zn, Cu, Mo, Fe and Mn, the mean values are 6.05 Zn, 1.60 Cu, 1.09 Mo. 4.61 Fe and 4.90 Mn respectively. Micronutrients though significantly different between samples, and lower when compared with control, all (including control) were at low levels at which deficiency can occur when compared with critical nutrient levels (table 3).*

***Keywords*— Cassava, Imo State, Mbaise, Micronutrient**.

I. INTRODUCTION

Decreasing soil fertility with declining yield growth for major food crops have raised concerns about the sustainability of agricultural production at current level (Mortvedt, 1995). . The work of Grundon (1997) added important reason for us to investigate nutrient management and elements levels in the soil. He posited that, we not only need food, but quality food too.This was supported by Paterson (2002) who posited that the deficiency of zinc for instance, to tuber crops, reduces the quality of carbohydrate in the tubers likewise nitrogen deficiency, which reduces protein content in grains (Havlin *et al*., 2006}. Many declines in crop yield have be associated with micronutrients levels (Enwezor *et al.*, 1990).

Micronutrients are elements or plant nutrients which are essential for plant growth, but are required in relatively small amounts than those of the primary nutrients (N,P,K,S, Mg etc).

These micronutrients include Iron (Fe), Copper (Cu), Manganese (Mn), zinc (Zn), Boron(B), Molybdenum(Mo) and Chloride(Cl) etc. Soil vary widely in their micronutrient content and in their ability to supply micronutrients in quantities sufficient for optimal crop growth (Solberg *et al.*, 1999). Micronutrients though required by plants in small quantity but their action in plant are not small (Wiese, 2010). And what quantity is sufficient to support plant for normal growth and development (Enwezor *et al.*, 1990).

Many studies (Jacobsen and Jasper, 1991; Akinrinde and Obigbesan, 2000) have revealed micronutrient deficiency in crops, and (Boardman and McGure, 1990) stressed that, micronutrients are important for plant growth as plants requires a proper balance of all the nutrients for normal growth and optimum yield. Excessive use and or, availability of these macronutrients over nonsufficient levels of micronutrients affects the soil pH, and this in turn, tend to decrease Manganese (Mn), Iron(Fe), Copper(Cu), and Boron(B) as pH increases (Alam and Raza, 2001).

Most farmers in this part of the country apply NPK fertilizer regularly with little or no attention to micronutrients, forgetting the danger of imbalances with micronutrient s(Jones, 2007). Findings from some researches in Nigeria have found micronutrient deficiencies in Nigerian soils (Mckenzie, 2003). Each deficiency symptom is related to some function of the nutrient in the plant Also, a considerable proportion of agricultural soils in Nigeria can be classified as low fertile soils (Bennett, 2003). To this end, there is need to investigate the levels of some micronutrients (Zn, Cu, Fe, Mo, Mn) on selected soils in the study area and also determine the physico-chemical properties (soil pH, organic matter content, cation exchange capacity, exchangeable acidity etc) of arable soils in the study area.

II. MATERIALS AND METHOD

2.1. Study Area

This study was carried out in four communities in Ezinihitte Mbaise LGA, Imo state. Ezinihitte Mbaise is strategically located in Imo State of Nigeria. Ezinihitte Mbaise has the following coordinate Latitude:5.50511, Longitude:7.36771 5° 30′ 18″ N, 7° 22′ 4″ E. The temperatures ranged from 32.1-29.1^{0c} (maximum) and 24.1-22.2^{0c} (minimum), while relative humidity in these areas ranged from 77-86%, while rainy season is from April- October with a short break in August Called “August Break”.

2.2 Site Selection, Description And Soil Sampling

Reconnaissance survey visits were made to locate and select sites for the study. The experiment was conducted in Ezinihitte Mbaise LGA. The experiment was carried out in four communities namely –Udo, Eziudo, Obizi and Onicha both in Ezinihitte Mbaise LGA in Imo state. The farmlands comprises of cassava continuous cropping. The control was uncultivated lands of about 5 years. The dominant trees present are oil palm tree, oil bean and the topography is undulating.

Representative soil sample were collected randomly. Sampling was done with the aid of soil auger at a depth of 30 cm since previous work by Nyanagababo and Hamya (1986) showed that surface soils are better indicator of trace elements concentrations. Samples were randomly collected (Brown, 1987) from the sites.

These 4 villages in Ezinihitte were sampled. 20 samples were collected from Udo, and 4 samples each were bucked for 4 composite replicates. The procedure was the same for all the other villages. The samples were emptied into a paper envelop respectively.

The control was sampled from 20 fallow lands, from each of these villages. Procedures previously discussed were adopted in replication.

2.3 SAMPLE PREPARATION AND LABORATORY ANALYSIS

2.3. 1 Soil Sample Analysis

Soil samples were spread on clean and dry paper sheet for air drying. After air drying, the samples were crushed in clean ceramic mortar using a small ceramic piston. These samples were passed through 2-mm sieve to get a fine soil fraction (Nelson and Sommers, 1982). The analyses was carried out at Federal College of land Resources and Technology (FECOLART) Oforola, Imo State.

The fine soil fraction was used to extract micronutrient using the DTPA method (Lindsay and Norvell, 1978). A 10 g of soil sample was mixed with 20 ml DTPA (0.05 M – adjusted to pH 7.3 with TEA), then shaken on a reciprocation shaker or (mechanical shaker) for 30 – 45 minutes before filtering through whatman No 1 filter.

The filtrate was analysed for micronutrients (Cu, Zn, Fe, Mo, Mn) on Atomic Absorption Spectrophotometer (AAS), Perkin Elmer model 306

Soil pH was determined in distilled (deionised) water (1:2.5 Soil-Water ratio) using glass electrode pH meter (Dewer model) as described by Smith and Doran(1996). **Organic carbon** was determined by the Walkley-Black wet oxidation method (Heanes, 1984).

Organic Matter was determined by multiplying organic carbon by 1.724(Bemmelen’s Factor). **Exchangeable**

acidity was determined by the titration method (McGrath and Cunliffe, 1985).

Cation Exchange Capacity was determined using summation method (McGrath and Cunliffe, 1985). **Particle Size Distribution** was determined using hydrometer method of mechanical Analysis. **Base Saturation** was calculated by dividing total exchangeable bases by effective cation exchange capacity value and multiplied by 100.**Total**

Table 1. Physicochemical property of the soil

Samples	Text ure	pH H_2O	Av. P Mg/kg	N ……………	OC %	OM …..................	BS	Ca ………………..	Mg	K Cmol.Kg-1	Na ……..……..……..	Ea	ECEC
Obizi (A)	LS	5.09^b	0.89^c	1.14^b	1.65^b	2.85^b	71.9^b	2.27^c	1.47b	0.17^a	0.11^a	1.60^b	5.61^b
Eziudo (B)	LS	5.44^a	2.52^a	1.11^b	1.29^c	2.23^c	78.7^a	2.53^b	1.53a	0.18^a	0.16^a	1.10^d	5.48^c
Onicha ©	LS	5.38^a	2.53^a	1.11^b	1.27^c	2.19^c	73.1^b	2.27^c	1.33c	0.18^a	0.16^a	1.43^c	5.39^c
Udo (D)	LS	5.15^b	1.61^b	1.10^b	1.21^c	2.09^d	70.4^b	2.40^b	1.60^a	0.11^a	0.08^b	1.47^c	4.95^d
Control €	LS	5.00^{bc}	0.32^d	2.20^a	2.61^a	4.59^a	77.6^a	2.86^a	1.46^b	0.13^a	0.16^a	1.80^a	5.96^a
Mean	**LS**	**5.21**	**1.57**	**1.33**	**1.60**	**2.79**	**74.3**	**2.46**	**1.47**	**0.15**	**0.13**	**1.48**	**5.47**

Within each column, means with different letters are significantly different at ($P < 0.05$).

Key: P phosphorus, N Nitrogen, OC organic carbon, Om Organic matter, BS Base Saturation, Ca calcium, Mg magnesium, k potassium, Na sodium, Ea Exchangeable acidity, ECEC Effective cation Exchange Acidity.

Nitrogen was determined using Kjeldahl method (Brown, 1987). **Available Phosphorus** was determined using Bray-2 method (Olson and sommer, 1982). **Effective Cation Exchange Capacity** was determined using summation method, that is, exchangeable base plus exchangeable acidic expressed in (molkg^{-1}).

2.4 Data Analysis

Simple descriptive technique was used and data were summed and divided to produce means respectively. Means were separated using the Least Significant Difference (LSD) according to Snedecor and Cochran (1980), correlation with physicochemical properties were made, and comparism were made with results from the control and already established levels.

III. RESULTS AND DISCUSSON

3.1.Some Mineral Nutrient Elements

The details of physical and chemical properties of the samples are shown in table 1.

3.1.1 Nitrogen, Phosphorus and Potassium –NPK

The total nitrogen level decreased significantly between treatments with the lowest occurring at Udo. Nitrogen levels were all lower when compared with control.

Phosphorus levels were also significant different when compared between samples and control. The samples were higher in available Phosphorus than control and the highest occurred in Onicha.

Potassium levels differ in all the samples and were significantly different between the samples, and when compared with control. With addiction of NPK fertilizers, levels of macronutrient increase tremendously and can decrease through plant uptake in actively growing fields occupied by crops and weeds (Marschner, 1995). For phosphorus, the levels were in accordance with Rengel (2001), who indicated that most Ultisols are sandy and possesses P fixing ability. P becomes available if moisture content is sufficient. The fertility levels of southeastern soils are moderate to low and soil amendments including fertilizer application is crucial especially under continues cultivation (MCkenzie, 2003).

3.1.2 Magnesium (Mg) and Calcium (Ca)

There were significant difference between the samples, and the lowest occurred in Onisha but were significantly different when compared with control.

Cultivation affected the calcium levels and there were significant different between some samples and these were significantly reduced when compared with control.

3.1.3 Sodium

Cultivation affected the sodium content of the sites slightly but significantly, and these were significant different between some samples Table 1. Moderately acid soils have Mg and Ca at moderate to high levels (Enwezor *et al.*, 1990).

3.2 Chemical Properties Of The Soil

The values of chemical properties were summarized in table 1.

3.2.1 Soil texture.

The soil texture is loamy sand. FMANR (1990) revealed that sandy soils are predominant in Imo State. This soil is always low is fertility indices and has higher leaching capacity as well as high drainage potential and low water and nutrient holding capacity.

3.2.2 pH

The soil pH were significantly different between samples and the lowest occurred in control sites. The soil is moderately acidic and this justifies the many findings that reported tropical soils as acidic (Akinrinde, *et al*., 2005). Cultivation increases soil acidity through applications of chemical fertilizers which further increase soil acidity (Akamigbo and Asadu, 2001).

3.2.3 Organic Matter Content (OM)

There were significant difference between samples and control when compared, and the highest occurred in Control.

The percentage organic carbon were also affected from one sample to another and were not significantly different between samples C and D except when compared with control. There is rapid decomposition of organic matter in tropical soils, and the levels are even lower in sandy soils (Brown, 1987). Soil organic matter help in nutrient retention and slow release. It increases soil water holding capacity and as source of energy for soil biota,

3.2.4 Percentage Base Saturation (% BS)

Percentage base saturation of the samples decreased slightly and significantly, and all except Eziudo (B) were lower than control.

3.2.5 Effective Cation Exchange Capacity (ECEC) and Exchangeable Acidity (EA)

Samples exchangeable acidity were all lower and significantly different from control. The ECEC of the sites were also affected. All the sites except control were lower significantly when compared. This levels were supported by Akamigbo and Asadu (2001) who posited that Imo State Soils are generally acidic and have moderate ECEC.

3.3 Micronutrient Levels

3.3 .1 Zinc

There were significant difference in zinc content of the sample, with highest concentration occurring at Onicha when compared with control. Numerous findings especially Enwezor *et al*. (1990) revealed that eastern soils are low in micronutrient except at municipal dump sites. **Zinc** is an essential component or various enzyme systems for energy production, protein synthesis as evidenced by accumulation of soluble nitrogen compounds such as amino acids and amides, and growth regulation. It has been conducted that zinc reduces auxin content through its involvement in the synthesis of tryptophan, a precursor of the auxin.

Deficiency of **zinc** results in light green, yellow or white areas between leaf veins, particularly in older leaves, premature foliage loss, malformation of fruits, often little or no yield, may occur. Zn in soil solution ranges from 2- 70 Mg/Kg with more than half complexed with organic matter. Deficiency of Zinc are usually associated with concentrations of less than 10- 20 Mg/Kg (Paterson, 2002). Depending on the crop, toxicity will occur when the leaf concentration of Zn exceeds 400 Mg/Kg.

3.3.2 Copper

Copper levels were lower in all sample when compared with control and were significantly different between samples. **Copper** is necessary for carbohydrate and nitrogen metabolism, legume synthesis which is needed for cell wall strength. It is also known to function in photosynthesis and respiration. Copper deficiency includes, die – back of stems, chlorosis, stunted growth, pale green leaves that wither easily. **Copper** deficiency and toxicity are not as common as other micronutrients deficiency. Copper deficiency include chlorosis in young leaves, and stunted. In advance stage, necrosis along leaf tips and edges appears. Stem melanosis, root rot and ergot infection can occur in small grains (Solberg *et al*., 1999). Cu toxicity include reduced shoot vigor, poorly developed and discolored root systems. Toxicity is uncommon, occurring where there are high deposits of waste such as municipal, sewage sludge etc. Concentration of Cu in soil ranges from 1- 40 Mg/Kg and averages about 9 Mg/Kg.

3.3.3 Molybdenum

The concentrations of molybdenum was lowest at eziudo, and highest in Udo. It was significantly different when compared with control which was almost at the same level with Onisha and Obizi. **Molybdenum** sufficient levels in the soil ranges from0-02-5mg/kg and is involved in enzyme systems relating to nitrogen fixation and metabolism, protein synthesis and sulphur metabolism, pollen fruit formation. Deficiency is not common but is similar to interveinal chlorosis in Iron deficiency. Excessive amount of Molybdenum are toxic, especially to grazing cattle or sheep. Mo toxicity cause stunted growth and bone deformation in animal and can be corrected by oral feeding of Copper (Paterson, 2002). The soil concentration of Mo ranges from 0.2 to 5 Mg/Kg (Benett, 2003).

3.3.4 Iron

There was a similar trend of concentrations in iron levels. There were significant differences in iron content and the highest occurring in control. Iron content of tropical soils are different from location to location due to parent materials (Osiname, 2005). **Iron** is involved in the production of chlorophyll, component of many enzymes for energy transfer, nitrogen reduction and fixation, lignin formation. It provides the electrochemical potentials for many enzymatic transformations in plants (Jones, 2007). **Iron** deficiency symptoms include interveinal chlorosis, which progresses rapidly over the entire leaf. In severe cases, leaves turn entirely white and necrotic (Van Dijk *et al*., 1993). Iron toxicity can occur under certain condition, for example in rice grown or poorly drained or submerged soils, leaf bronzing symptoms occur with 300 mg/kg of Fe in rice leaves. Fe concentration is usually very low, 0.1-0.50 Mg/Kg and only the chelate dynamics make Fe more available (Van Dijk *et al*., 1993). **Iron** is involved in the production of chlorophyll, component of many enzymes for energy transfer, nitrogen reduction and fixation, lignin formation. It provides the electrochemical potentials for many enzymatic transformations in plants (Jones, 2007).

Table 2:

Micronutrient levels of the samples

Source	Zn	Cu	Mo	Fe	Mn
	………………...Mg/Kg		………….………..		
Obizi (A)	3.12^{d}	2.19^{b}	0.07^{b}	2.75^{d}	1.34^{e}
Eziudo (B)	5.28^{c}	0.10^{cd}	0.03^{bc}	3.35^{e}	2.59^{d}
Onicha ©	6.59^{b}	2.18^{b}	0.07^{b}	5.80^{b}	7.33^{b}
Udo (D)	6.50^{b}	0.13^{c}	0.13^{a}	4.34^{c}	4.81^{c}
Control €	8.76^{a}	3.36^{a}	1.16^{a}	6.90^{a}	8.47^{a}
Mean	**6.05**	**1.60**	**1.09**	**4.61**	**4.90**

Within each column, means with different letters are significantly different at (P < 0.05)
Key: (Zn) Zinc, (Cu) Copper, (Mo), Molybdenum, (Fe) Iron, (Mn) Manganese.

3.3.5 Manganese Mn

~~Mn levels were significantly different and the highest~~ occurring in Onicha. These levels were in accordance with Adepetu (1990), who described Nigerian soils as lacking in Mn and other micronutrients. **Manganese** is necessary for photosynthesis, nitrogen metabolism and to form either compounds required for plant metabolism or enzyme activator. Manganese functions in nitrate reduction where it acts as an indicator for enzymes nitrate reductase and hydroxylamine reductase. **Its deficiency** results in interveinal chlorosis, brown necrotic spots appear on leaves, ~~premature leaf drop, delayed maturity, whitish grey spots~~ in leaves of cereals and shortened internodes in cotton. Manganese deficiency occur mainly in high pH soils, sandy soils low in organic matter and over limed soils (Gerendas, *et al*., 2009). Mn has a sufficient range of 15-100mg/kg.

Table 3. Critical levels of some nutrient Elements

Nutrient		**Low**	**Marginal**	**Sufficient**	**High**	**Excess**
	Spring	1.5	1.5 - 2.0	2.0 - 3.0	3.0 - 4.0	4.0
Nitrogen (N) %	Winter	1.25	1.25 - 1.75	1.75 - 3.0	3.0 - 4.0	4.0
Phosphorous (P) %		0.15	0.15 - 0.25	0.26 - 0.5	0.5 - 0.8	0.8
Potassium (K) %		1.0	1.0 - 1.5	1.5 - 3.0	3.0 - 5.0	5.0
Sulphur (S) %		0.1	0.1 - 0.15	0.15 - 0.40	0.40 - 0.8	0.8
Calcium (Ca) %	Other	0.10	0.10 - 0.2	0.2 - 1.0	1.0 - 1.5	1.5
Magnesium (Mg) %		0.1	0.1 - 0.15	0.15 - 0.50	0.5 - 1.0	1.0
Zinc (Zn) mg/kg		10	10 - 15	15 - 70	70 - 150	150
Copper (Cu) mg/kg	Barley	2.3	2.3 - 3.7	3.7 - 25	25 - 50	50
Iron (Fe) mg/kg		15	15 - 20	20 - 250	250 - 500	500
Manganese (Mn) mg/kg		10	10 - 15	15 - 100	100 - 250	250
Boron (B) mg/kg		3	3 - 5	5 - 25	25 - 75	75
Molybdenum (Mo) mg/kg		0.01	.01 - .02	.03 - 5	5 - 10	10

Sources: Westerman (1990).

IV. CONCLUSION

The result of the study showed that micronutrient contents are generally low when compared with control. Some are at deficiency zone at which deficiency can occur. And for normal plant growth and development, and also for optimum yield, these levels can totally affect yield critically. There were more positive correlations between the micronutrients and some selected samples and parameters. These also indicated that micronutrient was affected directly by cultivation. This reminds us that, in as much as these nutrient elements are needed in small quantity (ies); what quantity is good enough for optimum crop yield should be taken into consideration. Also, what quantity (ies) guarantees quality yield should not be ignored because quality of produce is better than quantity in nutritional terms? In targeting to get quality produce, quantity is guaranteed. It should be wrong if we measure out produce with quantity alone. In a scientific word, both quantity and quality should be a policy statement to guide the farmers. Most importantly, some micronutrients are directly involved in plant growth and developments, thus, it will amount to waste of human resources if we allow nutrient deficiency to result to low yield or in serious cases, crop failure. Deficiency of zinc for instance, to tuber crops, reduces the quality of carbohydrate in the tubers likewise nitrogen deficiency, which reduces protein content in grains. The results suggests addition of micronutrients to soils during cultivation because the micronutrient levels were below critical(sufficient) (table 3) levels and this cannot guarantee optimum yield taken cognizance of functions of micronutrients in cassava and crop production.

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Potentials of Soil on Palm Kernel Oil Free Fatty Acid

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Abstract*— Potentials of soil on Free Fatty Acid from Palm kernel Oil was conducted to correlate the soil physiochemical parameters and heavy metals to the free fatty acid of the palm kernel oil. Five samples of the soil were collected from Mbano and a sample of locally made palm kernel oil. The parameters analyzed were pH, temperature, organic matter, moisture, electrical conductivity, heavy Metals and free fatty acid of the oil. Results revealed : pH (5.57 ±0.49, 5.64 ± 0.36, 6.14 ±0.07, 5.95 ±0.16, 6.79±0.47), temperature (2.81 ±1.06, 28.0 ±0.83, 29.7 ±0.57, 29.7 ±0.64, 28.2 ±1.06), moisture (11.0 ±0.1, 18.0 ±0.2, 11.9 ±0.15, 12.9 ±0.25, 17.4 ±0.7), electrical conductivity (624 ±0.1, 541 ±0.2, 482.6 ±0.2, 477.0 ±0.1, 619.6 ±0.2) organic matter (77.7 ±0.15, 94.08 ±0.02, 48.6 ±0.25, 68.2 ±0.25, 38.8 ±0.15) Ca (0.644, 3.235, 1.453, 1.015, 1.673), Mg (1.467, 0.892, 1.483, 1.687, 1.147), K (2.017, 3.0054, 2.013, 1.0028, 2.067), Pb (0.036, 0.017, 0.015, 0.00, 0.0017), Fe (0.416, 0.826, 0.318, 0.316, 0.544), Na (0.656, 1.450, 0.908, 1.751, 1.956) cation exchange capacity (4.78, 8.58, 5.83, 5.45, 6.84) and free fatty acid (2.7 ±0.15, 4.3 ±0.15, 6.1 ±0.1,7.6 ±0.15,11.4 ±0.2). There was a strong positive correlation between the free fatty acid of palm kernel oil and soil sodium concentration. The study revealed that quality of the soil has a relationship with the free fatty acid of the palm oil.

***Keyword*— PKO, Potentials, freefatty, Acid, soil**

I. INTRODUCTION

Palm Kernel Oil is a triglyceride typically unctuous, viscous, combustible, liquid at ordinary temperatures, and soluble in ether or alcohol but not in water [1,2].

The triesters of fatty acids with glycerol (1,2,3-trihydroxypropane) compose the class of lipids known as fats and oils. These triglycerides (or triacyglycerols) are found in both plants and animals, and compose one of the major food groups of our diet. Triglycerides that are solid or semi-solid at room temperature are classified as fats and oils, and occur predominantly in animals [3]. Those triglycerides that are liquid are called oils and originate chiefly in plants. Fats have a predominance of saturated fatty acids, and oils are composed largely of unsaturated acids [4].

Soil is the mixture of minerals, organic matter, gases, liquids and countless organisms that together support life on earth [5]. Soil is the mixture of minerals, organic matter, gases, liquids, and the countless organisms that together support life on earth. [6] Soil is a natural body known as the pedosphere and which performs four important functions: it is a medium for plant growth; it is a means of water storage, supply and purification; it is a modifier of the atmosphere of earth: it is a habitat for organisms all of which, in turn, modify the soil [7,8]. Soil is considered to be the "skin of the earth" and interfaces with its lithosphere and biosphere· Soil is consists of a solid phase (minerals and organic matter) as well as a porous phase that holds gases and water ·It is often treated as a three state system [9,10].

The chemistry of a soil determines its ability to supply available plant nutrients and affects its physical properties and the health of its microbial population [11].

The aim of the research was to correlate the physiochemical parameters and heavy metals in soil to free fatty acid in palm kernel oil in other to determine the trend To achieve this aim specific objectives were to determine the physiochemical parameters of soil and oil, determine the some heavy metals content in soil and determine the relationship between physicochemical parameters and metals in soil and free fatty acid in palm kernel oil.

II. MATERIALS AND METHOD

2.1 Sampling Sites

Sampling sites were selected to reflect spatial variability and quality of the soil associated with sampling zone. The locations includes- Anara – Sample A, Obollo –Sample B, Amauzari – Sample C. Osuachara - Sample D, Oka – Sample E

2.2 Sampling and Pretreatment

Twenty five samples were collected from the five different sites. At each site a **W** shaped line was drawn on a 2×2m surface along which five samples were collected from each 0f the top soil area and mixed homogeneously to form one sample [12, 13]. The soil samples were taken from 0-15cm depth. The soil samples were five samples in number and stored in a polyethene bag and taken to Imo State University laboratory for analysis.

2.3 Visual Classification

The colour was determined with the naked eyes and a standard munsell colour chart. The soil sample was placed on a paper to detect the moisture effect. This was determined using a soil textural triangle. Texture Group was determined by felling the soil sample by hand while Odour was was determined by smelling the sample. Evidence of Contamination was assessed when The soil sample was placed on a white cloth, which it stained [14,15,16].

2.4 Quality assurance, Chemicals and reagent

All instrument used in this work were in good working condition and were used according to manufacturer's instructions. Aqua-Regia, Hydrochloric acid (HCl), Nitric acid (HNO3), Ethanol,Sodium hydroxide (NaOH) (0.1N), Phenolphthalein indicator, Potassium Chloride (KCl), Distilled water

2.5 Determination of Physicochemical properties

50g of soil sample was weighed and poured into an empty, clean quart jar. Water was added in the quart jar and left overnight. The sand, silt, and clay was examined and the soil texture was determined using a soil textural triangle [17].

Reaction With Hydrochloric Acid was done as follows: 10ml 0f Hydrochloric Acid and 30ml of Distilled water was measured. Dilute the Hydochloric Acid by pouring it into distilled water. 5g of of soil sample was weighed. Place each sample on a filter paper; the Hydrochloric Acid was gradually poured on the soil sample to examine the reaction. The P^{H} values of the soil samples were determined using a Jenway 3510 PH meter. The pH was determined by dipping the electrode into a 2:1 soil/water mixture that had been stirred and allowed to equilibrate for about one hour. The pH meter was calibrated with pH 7.0 and pH 4.0 buffer before use. Electrical conductivity was determined using Hanna (HI 8733).Half of a cup of the dried soil was measured and poured into a beaker, half of a cup of distilled water was added into the beaker and the mixture was stirred gently for 30 seconds. The soil - water suspension was allowed to stand for 30minutes and stirred again [18, 19, 20].

The probe of the Electrical conductivity was inserted into the soil solution and swirled gently in the soil – water extract .Conductivity was determined after about 30 – 60 seconds when the Electrical conductivity meter has stabilized [21]·The Electrical conductivity meter was calibrated with potassium chloride solution before use.

The temperature of the soil sample was determined using a Gardener soil thermometer. The soil sample was poured into a quart jar and the thermometer bulb was inserted into a 3cm depth of the soil inside the quart jar [22].

An Oven (Drier Box DHG-9053) was use in drying the soil sample.10g of the soil sample was weighed into a porcelain dish and dried in the oven for 24 hours at 106^{O}C. The dry sample was reweighed to determine how much water was lost.

The organic matter of the soil sample was determined by recording the mass of an empty, clean and dry porcelain dish (MP). The entire oven – dried test specimen from the moisture content experiment was placed in the porcelain dish and the mass recorded (MPDS). The dish was placed in a muffle furnance at 248ºC and left in the furnance overnight. The porcelain dish was removed using the tongs and alow to cool to room temperature .The mass of the dish containing the ash (burned soil) was recorded [23,24].

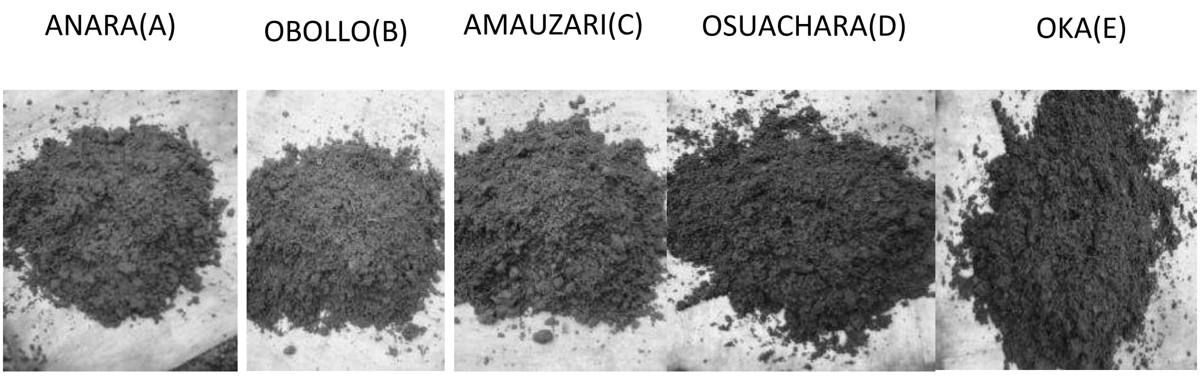
2.6 Determination Of Heavy Metals

The heavy metals was determined using Atomic Absorption Spectrometer(VARIAN AA 240). 1g of the soil sample was weighed into a test tube and digested with 24ml of Aqua Regia then left for two days. The mixture was filtered and the filtrate was poured into a sampling container, then analyzed in the Atomic Absorption Spectometer [25].

2.7 Sampling and Analysis of Palm Oil

Sampling Sites

Three oil samples were bought from Orie Amaraku. The oil was checked for adulteration.



Figs 1. : Photos of soil samples on paper

2.7.1 Physiochemical Parameters of oil

2.7.2 Determination of Free Fatty Acid

5g of palm kernel oil was measured into a clean dry conical flask and make up to 100 cm3 of ethanol, and then heat on water bath till boiling. 2 -3 drops of phenolphthalein was added and the mixture was mixed properly. Titrate with 0.1 m NaoH, shake vigorously till the appearance of a pink colour, which persist for at least 30 seconds. Measure the volume of sodium hydroxide titrant [26, 27].

2.7.3 Determination of Moisture Content

The moisture content was determined using oven (drier box dhg -9053). 10g of palm kernel oil was weighed and poured into a known weight of an empty beaker. The oil in the beaker was kept in an oven for 6 hours and maintained at a temperature of 105^{O}C, allowed to cool and reweighed to a constant weight [28].

Table 1: Visual Characteristics of soil samples from Mbano

Classification	AnaraA)	Obollo (B)	Amauzari(C)	Osuachara (D)	Oka(E)
Colour	Brown	Dark brown	Brown	Brown	Black
Moisture description	Moist	Moist	Moist	Moist	Moist
Texture group	Medium	Medium	Medium	Medium	Medium
Texture class	Sandy loam	Sandy loam	Sandy loam	Sandy loam	Sandy loam
Odour	Organic	Organic	Organic	Organic	Organic
Evidence of contamination	Staining	Staining	Staining	Staining	Staining
Other comment	Presence of living organism and debris	Presence of living organism and debris	Presence of living organism and debris	Presence of living organism and debris	Presence of living organism and debris

2.7.3 Determination of Viscosity

The viscosity of the oil was determined using a Rotary viscometer Test method. The oil was placed in a beaker, housed in an insulated block at a fixed temperature. A metal spindle is then rotated in the oil at a fixed rpm, and the torque required to rotate the spindle is measured [29].

2.7.4 Determination of pH

The pH value of the oil was determined using a Jenway 3510 pH meter. The oil was poured into a 200ml beaker; the pH electrode was dipped into the oil. The pH meter was calibrated with pH 7.0 and 4.0 buffer before use [30].

III. RESULTS AND DISCUSSION

The results of the visual characteristics, physiochemical parameters, heavy metals of the soil were summarize in table 3.1, 3.2, and 3.3

3.1 Visual Characteristics

In table1, sample A,B,C,D has a Dark Brown, Brown colour and sample E a Black colour which indicates the presence of organic matter and decaying vegetation which make the soil sample fertile and stores plant nutrients.The texture class and group which is a medium sandy loam soil and has organic odour because of the presence of organic matter in it [31,32].

3.2 Physiochemical Parameters

From graph 1 in figure 2: There is a strong positive correlation between the free fatty acid of palm kernel oil and the pH, (R =0.9466) and (R^2= 0.8961) .As the pH increases the free fatty acid increases , the relationship between the variable is strong. From graph 2 in figure 3, There is a positive correlation between the free fatty acid of palm kernel oil and the temperature, (R =0.1776) and (R^2=0.0315), but the relationship between the variables is weak. The temperature don't have much effect on the free fatty acid. From graph 3 in figure 4: A positive correlation exist between the free fatty acid of palm kernel oil and the moisture content,(R= 0.4391) and (R^2=0.1928), the relationship between the variables is weak. The moisture content has little effect on the free fatty acid. From graph 4 in figure 5: There is a positive correlation between the free fatty acid of palm kernel oil and the Electrical conductivity, but has a weak relationship. (R=0.0261) and (R^2=0.0007).Electrical conductivity has little effect on the free fatty acid .

Table 2: Physiochemical parameters and OM

SAMPLES	Texture	pH	Temp (^OC)	Moist (%)	EC (μS)	RXN with HCl	OM (%)
Anara (A)	Sandy	5.57 ±0.49	28.1 ± 1.06	11.0 ± 0.1	624± 0.1	Strong	77.7±0.15
Obollo (B)	Sandy	5.64 ± 0.36	28.0 ± 0.83	18.0 ± 0.2	541.5 ± 0.2	S.strong	94.08±0.02
Amauzari (C)	Sandy	6.14 ±0.07	29.7 ± 0.57	11.9 ±0.15	482.6 ±0.2	S.strong	48.6±0.25
Osuachara (D)	Sandy	5.95 ±0.16	29.7 ±0.64	12.9 ±0.25	477.0 ±0.1	Strong	68.2±0.25
Oka (E)	Sandy	6.79 ±0.47	28.2 ±1.06	17.4 ±0.7	619.6 ±0.2	Strong	38.8±0.15

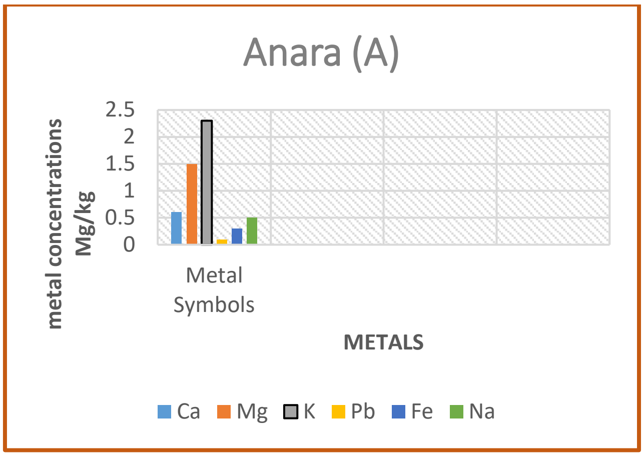
From graph 5 in figure 6: This has a moderate negative correlation between the free fatty acid of palm kernel oil and the organic matter,(R =-0.6379) and (R^2=0.4069) which means that as the free fatty acid increases the organic matter decrease or as the organic matter increases the free fatty acid decreases. Organic matter has an opposite effect on the free fatty acid [33, 34, 35, 36, 37].

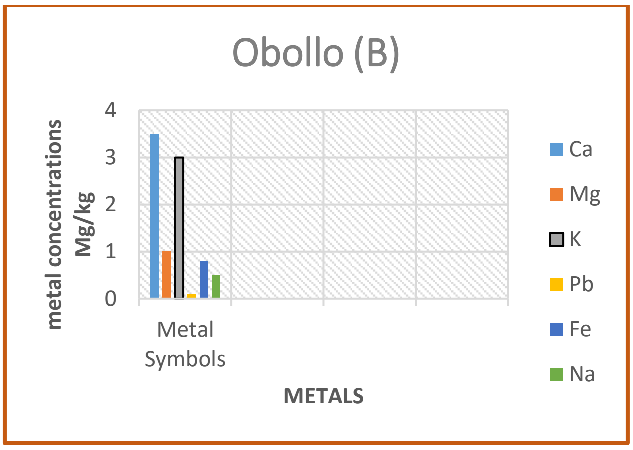
3.3 Heavy Metals in Soil

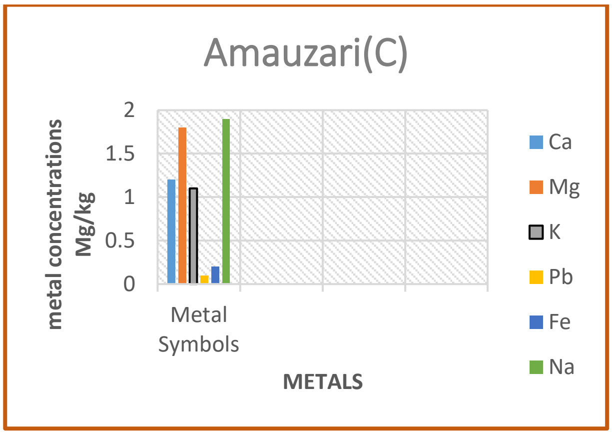
From graph 6 in figure 7: There is a positive correlation coefficient between the free fatty acid of palm kernel oil and the cation exchange capacity, (R=0.09) and (R^2 =0.0081). It has a weak relationship. The cation exchange capacity has little effect on the free fatty acid .From graph 7 in figure 8: There is a negative correlation between the free fatty acid of palm kernel oil and calcium, (R= -0.0139) and (R^2= 0.0002). It has a weak relationship. Calcium has little effect on free fatty acid. From graph 8 in figure 9: A negative correlation exist between the free fatty acid of palm kernel oil and magnesium(R=-0.0316) and (R^2 = 0.001).It has a weak relationship. Magnesium has a little effect on the free fatty acid. From graph 9 in figure 10: A negative correlation exist between the free fatty acid of palm kernel oil and potassium,(R =-0.3217) and (R^2 =0.1035). It has a weak relationship. Potassium has a little effect on the free fatty acid. From graph 10 in figure 11: A moderate negative correlation exist between the free fatty acid of palm kernel oil and lead, (R=-0.7325) and (R^2= 0.5366).As free fatty acid increases it goes with a decrease in lead, or as lead increases the free fatty acid decreases. From graph 11 in figure 12: There is a negative correlation between the free fatty acid of palm kernel oil and iron,(R=-0.1503) and (R^2= 0.0226).it has a weak relationship. iron has a little effect on the free fatty acid. From graph 12 in figure 13: There is a strong positive correlation between the free fatty acid of palm kernel oil and sodium, (R=0.8119) and (R^2= 0.6592).This means as the sodium increases it goes with an increase in the free fatty acid and as the free fatty acid increase it goes with an increase in sodium [37,38,39].

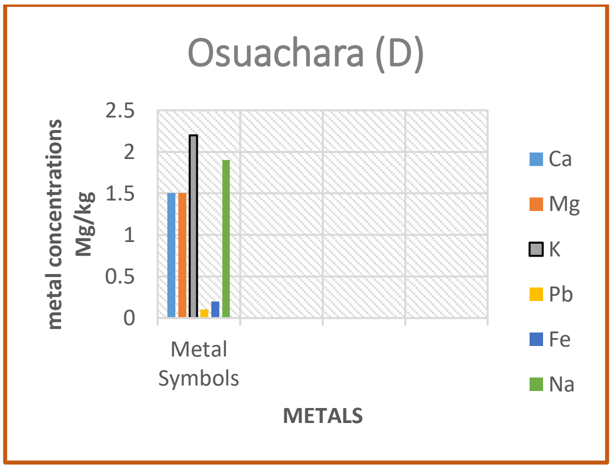
Table 3: Mean Heavy Metals concentrations (mg/kg) of soils at various locations

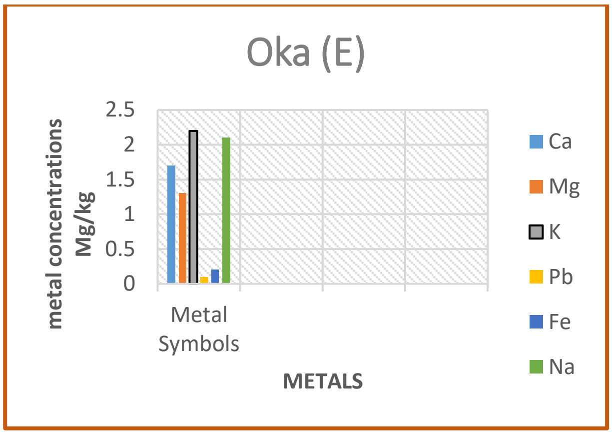
SAMPLES	Ca	Mg	K	Pb	Fe	Na	CEC
ANARA (A)	0.644	1.467	2.017	0.036	0.416	0.656	4.78
OBOLLO(B)	3.235	0.892	3.0054	0.017	0.826	1.450	8.58
AMAUZARI (C)	1.453	1.483	2.013	0.015	0.318	0.908	5.83
OSUACHARA (D)	1.015	1.687	1.0028	0.00	0.136	1.751	5.45
OKA (E)	1.673	1.147	2.067	0.017	0.544	1.956	6.84



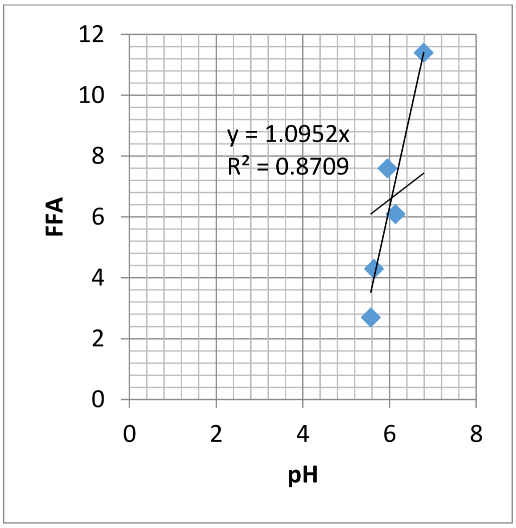


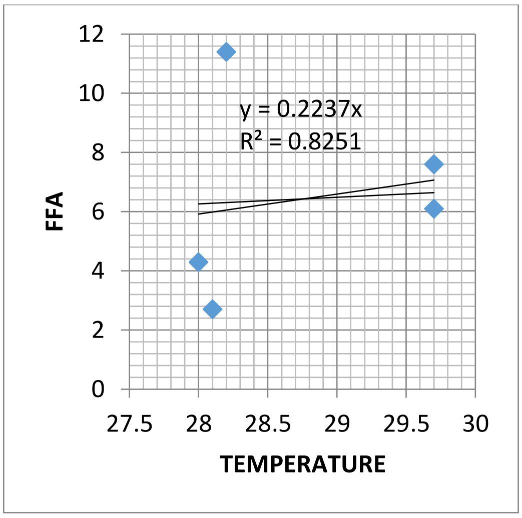


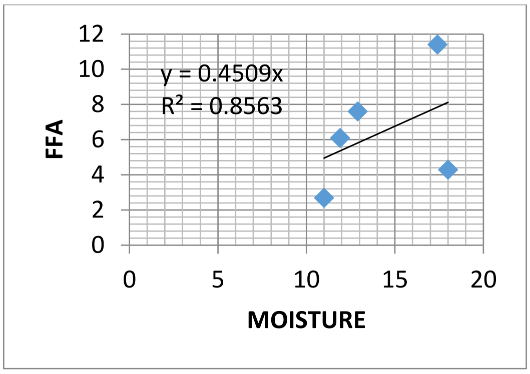


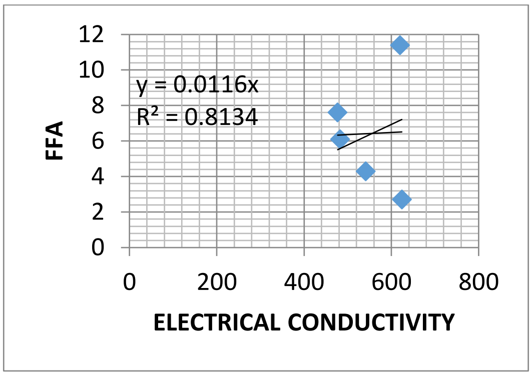


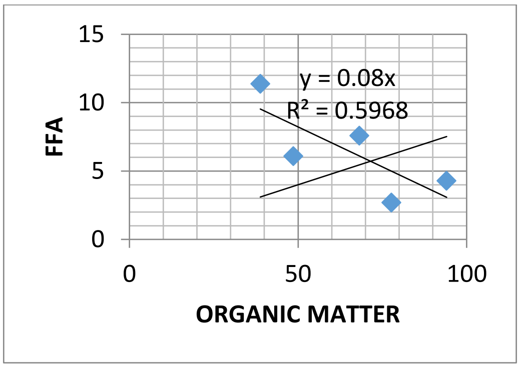
3.4 Correlation of physiochemical parameters in soil to free fatty acid in oil

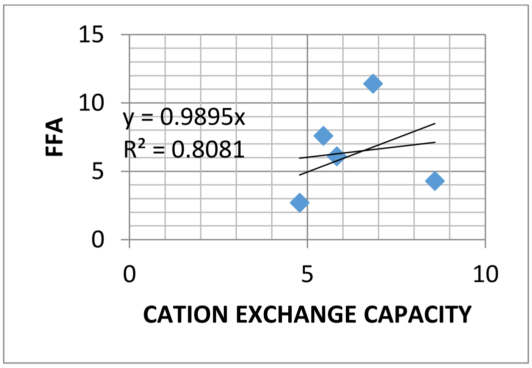


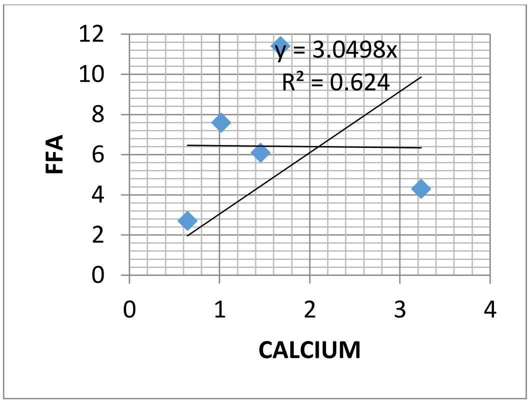


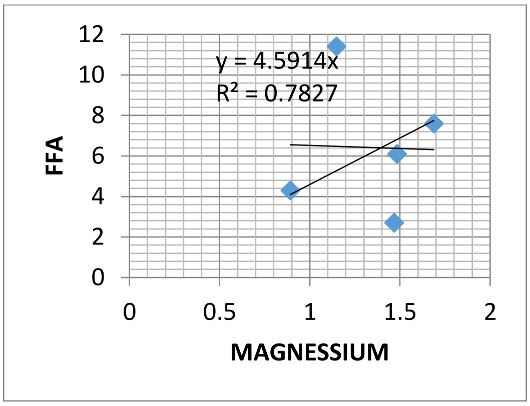


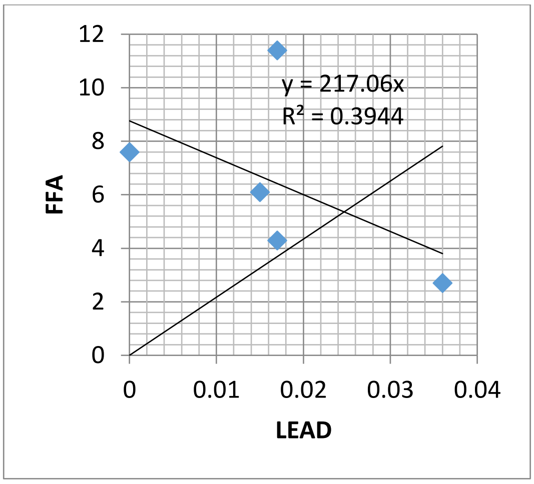












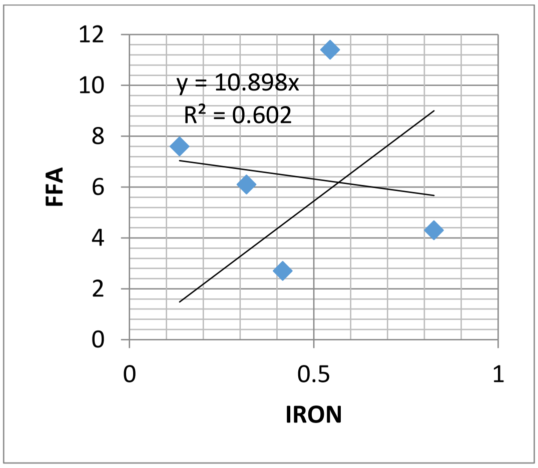


Fig.6- 12: Correlation of physiochemical parameters of soil and free fatty acid in oil

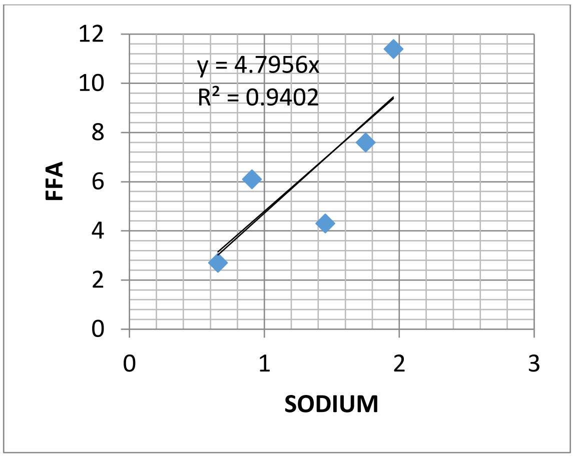


Fig 13-16: Correlation of metals in soil and free fatty acid in oil

3.5 Physiochemical parameters of oil

The pH (5.28 ± 0.01) is indicative of the presence of fatty acids in the oil, which is a good indicator of the advantageous utilization of the oil as a result of the presence of free fatty acids [40].

IV. CONCLUSION

The physiochemical parameters and heavy metals of the soil showed a positive and negative correlation with the free fatty acid of the palm oil. Therefore soil quality have an effect on the free fatty acid of the oil. However there was no defined trend of the relationship between soil and oil properties.

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Sustainable Airport Water Management: The Case of Hong Kong International Airport

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Abstract— *Utilizing an in-depth longitudinal case study research design, this study has examined Hong Kong International Airport's water management and the annual trends in the airport's water consumption for the study period of 2011 to 2020. Since its inception of operations in 1998, the airport has used a "triple water system", that has been designed to improve the efficiency of its three major water sources: freshwater, seawater and treated wastewater. The largest water source used by the airport is seawater. In the early years of the study (2011 to 2013), there was a general upward trend in the airport's annual municipal supplied water consumption. However, from 2014 to 2020, there was an overall downward trend in the airports municipal water consumption at the airport. The municipal supplied water consumption per enplaned passenger or per workload unit (WLU) largely displayed a general downward trend, which is very favorable given the strong growth in enplaned passengers recorded during the study period. The airport's annual seawater consumption, annual recycled/re-used water consumption and the annual discharged waters oscillated over the study period reflecting differing water consumption patterns.*

Keywords— ***Airport water management; Case study; Hong Kong International Airport; Water consumption, Water recycling/re-use***

I. INTRODUCTION

Airports are one of the critical actors in the global air transport value chain, as they provide the necessary ground-based and airfield-related infrastructure that facilitates the movement of passengers and air cargo between the air and surface transport modes. However, in delivering air transportation services, the key actors in the global air transport value chain require water, which is used in the provision of their services and for maintaining their infrastructure and equipment. To facilitate the provision of these aviation-related services, airports require and use very substantial amounts of water (Fossi & Esposito, 2015). Airports principally source waters from municipal authorities. Other sources of water are from rainwater harvesting (Somerville et al., 2015), and from seawater. Hong Kong International Airport, the case airport in this study, uses seawater as one of its key water sources. In addition, airports can produce very significant volumes of storm or waste waters (Baxter, 2022). These waters need to be handled in an environmentally sustainable way to mitigate their potential adverse impact on the environment. Thus, stormwater runoff at airports is a significant issue for airport operators (Shi et al., 2017).

Sustainable water management has increasingly become a very important element in airport's sustainability policies and, as a result, airports are increasingly focusing on ways to mitigate their water consumption. This is because airports consume large amounts of water. Airports are also focusing on the safe and environmentally friendly management of waste waters (Baxter, 2022; Pitt et al., 2002).

One such airport that has sustainably managed its water requirements throughout its inception of operations in 1998 is Hong Kong International Airport. Hong Kong International Airport is one of the world's major global passenger and air cargo hubs (Govada et al., 2017). In addition, Hong Kong International Airport (HKIA) is one of the principal gateways to Mainland China and is the major aviation hub in Asia (Tsui et al., 2014; Yuen et al., 2020). The mainland China market is Hong Kong International Airport's (HKIA's) largest air travel market (Centre for Aviation, 2019).

The objective of this study is to empirically examine how Hong Kong International Airport sustainably manages its water consumption and the discharge of its wastewaters. Hong Kong International Airport was selected as the case airport due to its long held sustainable water management practices. A second objective of the study was to examine how the increases in passenger traffic recorded at the airport have influenced the airport's water consumption during the study period. Hong Kong International Airport was selected as the case airport as it is a hub airport that is served by both full-service network carriers (FSNCs), low-cost carriers (LCCs), regional airlines, and dedicated air cargo carrying airlines. The availability of a comprehensive data set covering the period 2011 to 2020, was a further factor in selecting Hong Kong International Airport as the study's selected case airport.

The remainder of the paper is organized as follows: The literature review that sets the context of the case study is presented in Section 2. The research method that underpinned the case study is presented in Section 3. The Hong Kong International Airport case study is presented in Section 4. Section 5 presents the findings of the study.

II. BACKGROUND

2.1. Airport Water Management

The water consumption of airports is very substantial, as airports and their key stakeholders require large amounts of water to maintain their infrastructure and to support and facilitate their operational activities (de Castro Carvalho et al., 2013). Airports are also the source of run-off waters (Baxter et al., 2018b; Sulej et al., 2011; Sulej-Suchomska et al., 2016). Due to the significant impact that the high-water consumption and the runoff waters have on the environment, airports are now placing a very high focus on sustainable water management (Somerville et al, 2015). Indeed, sustainable water management is now a key element in many airports environmental and sustainability policies and practices (Baxter, 2021).

2.2. Airport Stakeholders Water Requirements and Usage

As previously noted, airports consume substantial volumes of water to maintain both their infrastructure, and thus, sustain their aircraft and ground-based operations (Baxter et al., 2018a; de Castro Carvalho et al., 2013, Neto et al., 2012). Airport operators, airlines, air traffic management agencies, ground handling agents, aircraft, and ground service equipment (GSE) maintenance organizations, airport concessionaires, and passengers and staff require water for drinking, catering, retail, cleaning, flushing toilets, and system maintenance. Water is also used to maintain an airport grounds and during the landscaping of gardens and parks that are located within the airport precinct (Thomas & Hooper, 2013).

2.3. Airport Water Sources

Historically, airports were designed to make use of ground water or water supplied from municipal authorities that satisfy appropriate quality standards. Where this water has only been used for non-industrial purposes (for example, washing, cleaning, and laundry), wastewater can be collected by the airport, treated, and reused for activities including toilet flushing, washing, and in some instances irrigation of plants. Such practices may require the airport to introduce a dual drainage system as well as water-purification facilities (Thomas & Hooper, 2013).

A further source of water comes from harvesting (collecting) and storing rainwater (Abdulla et al., 2021; Somerville et al., 2015; Yannopoulis et al., 2019). If implemented at an airport, then rainwater harvesting can substantially reduce the volume of water sourced from conventional supplies and acts as a reservoir to guard against water shortages. The most sustainable approach to water management is for airports seeking to become self-sufficient in their water supply by optimizing opportunities for water harvesting, recycling, and reducing consumption (Thomas & Hooper, 2013).

2.4. Airport Runoff Waters

In an airport's operational area, run-off waters can have a very serious environmental threat. These waters could have a negative impact on both soil and groundwater since they contain a relatively high concentration of contaminants (Vanker et al., 2013). Wastes associated with aircraft refueling, aircraft operations, aircraft, and ground service equipment (GSE) maintenance and equipment and facilities cleaning can potentially enter lakes and streams located nearby to the airport via the airport's storm water drainage system. Major aircraft overhauls that use toxic chemicals to remove paint can also pose a significant environmental threat should these toxic chemicals enter the water system (Culberson, 2011). Other contaminants originating from other airport operations or activities include detergent formulations, solids, oils, greases, residues, solvent residues, and heavy metals (Grantham, 1996). The discharge of fire-fighting foam during an aircraft emergency (Fawell, 2014) together with the production of in-flight meals, and meals served at restaurants and staff canteens also contribute grease and detergents to the wastewater generated at an airport (Baxter et al., 2018b).

2.5. Airport Water Processing Plants

Rainwater from the paved areas, particularly from the airport's apron areas, can be cleaned using a special treatment plant located at the airport. This facility will separate oil products from the waters. Alternatively, a collector can be connected to the local municipal treatment plant. Fuel storage, and aircraft hangars and aircraft and ground service equipment (GSE) maintenance facilities, should be equipped with traps to catch any waste oil products. These facilities should be inspected regularly (Kazda et al., 2015).

2.6. Airport Water Conservation Measures

Because of the increasing pressure to reduce water consumption and conserve available water resources, airports can implement a range of measures that will enable them to reduce their water consumption. To achieve their environmental-related and sustainability goals, many airports around the world have implemented a range of water conservation measures (Dimitriou & Voskaki, 2011). These water conservation measures include the overall reduction in water consumption at the airport (Baxter et al., 2018a, 2018b; Rossi & Cancelliere, 2013), re-using water from the treatment of waters at wastewater and sewage treatment plants in toilet facilities and for irrigation purposes (Baxter et al., 2018b; Chen et al., 2012), using rainwater for the flushing of the toilets in airport buildings and facilities (Baxter et al., 2018a; Yu et al., 2013), protecting groundwater from pollution (Gupta & Onta, 1997), the overall monitoring of water consumption at the airport (Boyle et al., 2013), and monitoring the surface and ground water quality (Bartram & Balance, 1996; Baxter et al., 2018b; Thomas & Hooper, 2013). Airports also need to protect surface and ground water resources (Thomas & Hooper, 2013).

III. RESEARCH METHODOLOGY

3.1 Research Method

The study's qualitative analysis was underpinned by a longitudinal case study research design (Derrington, 2019; Hassett & Paavilainen-Mäntymäki, 2013; Neale, 2019). The primary advantage of a qualitative longitudinal research design is that it reveals change and growth in an outcome over time (Kalaian & Kasim, 2008). A case study allows for the in-depth examination of complex phenomena (Cua & Garrett, 2009; Remenyi et al., 2010; Yin 2018). Case studies also permit researchers to gather and analyze rich, explanatory information (Ang, 2014; Mentzer & Flint, 1997). A further advantage of case studies is that they enable researchers to build theory and connect with practice (McCutchen & Meredith, 1993).

3.2 Data Collection

The qualitative data was sourced from the Airport Authority of Hong Kong annual sustainability reports. Thus, in this study secondary data was used to investigate the research objectives. The three guiding principles of data collection in case study research as recommended by Yin (2018) were followed in this study: the use of multiple sources of case evidence, creation of a database on the subject, and the establishment of a chain of evidence.

3.3 Data Analysis

The data collected for the case studies was examined using document analysis. Document analysis is often used in case studies and focuses on the information and data from formal documents and company records (Ramon Gil-Garcia, 2012). Existing documents provide a vital source of qualitative data (Woods & Graber, 2017). Furthermore, documents are one of the principal forms of data sources for the interpretation and analysis in case study research (Olson, 2010). The documents collected for the present study were examined according to four criteria: authenticity, credibility, representativeness, and meaning (Fitzgerald, 2012; Scott, 2004, 2014).

The key words used in the database searches included "Airport Authority of Hong Kong environmental management policy", "Hong Kong International Airport's principal water sources", "Hong Kong International Airport's total annual water consumption", "Hong Kong International Airport's total annual municipal supplied water consumption"; "Hong Kong International Airport's total annual seawater consumption"; "Hong Kong International Airport's total annual water consumption per passenger", "Hong Kong International Airport's total annual recycled/reused waters", and "Hong Kong International Airport's total annual discharged waters".

The study's document analysis was conducted in six distinct phases. The first phase involved planning the types and required documentation and ascertaining their availability for the study. In the second phase, the data collection involved sourcing the documents from Fraport AG and developing and implementing a scheme for managing the gathered documents. The collected documents were examined to assess their authenticity, credibility and to identify any potential bias in the third phase of the document analysis process. In the fourth phase, the content of the collected documents was carefully examined, and the key themes and issues were identified and recorded. The fifth phase involved the deliberation and refinement to identify any difficulties associated with the documents, reviewing sources, as well as exploring the documents content. In the sixth and final phase, the analysis of the data was completed (O'Leary 2004).

In this study, all the gathered documents were downloaded and stored in a case study database (Yin 2018). The documents were all in English. Each document was carefully read, and key themes were coded and recorded (Baxter, 2022).

IV. RESULTS

4.1. An Overview of Hong Kong International Airport

The Hong Kong Government announced their intention to construct a new international airport at Chek Lap Kok with the new airport replacing the country's Kai Tak Airport, which was capacity restrained. Chek Lap Kok Airport is located off the north coast of Lantau Island which is itself located off the west coast of the Kowloon peninsula and to the northwest of Hong Kong Island (Staddon & Fan, 1994).

Hong Kong's Chek Lap Kok International Airport was constructed through the reclamation of two small islands, namely Chek Lap Kok and Lam Chau. These islands are located 25 kilometres west of Hong Kong Island, and this served as the base for the new 1,255-hectare airport site (Chow et al., 2004). The construction of the new airport involved land reclamation from the surrounding water (Wu et al., 2020). The new Hong Kong Airport at Chek Lap Kok was constructed on a 1,248-hectare offshore reclamation platform (Covil & James, 1997; Pickles & Tosen, 1998; Plant, 1997), whilst a total area of 928 hectares was from reclaimed land (Berner, 1998; Endicott & Fraser, 2001). Hong Kong's airport relocated from Kai Tak to Chek Lap Kok Airport (Hong Kong International Airport) in 1998 (Kwong & Miscevic, 2002; Li & Loo, 2016; Zheng et al., 2020). In addition to the construction of a brand-new airport that had the capacity to handle up to 87 million passengers and 9 million tonnes of air cargo a year (Tsang, 1998). The airport occupies a site of 1,255 hectares (Airport Authority of Hong Kong, 2021b).

Air services are provided from Hong Kong International Airport to more than 190 cities around the world, including 50 Mainland China destinations (Choi, 2019). Hong Kong International Airport is the home base for Air Hong Kong, Cathay Pacific Airways and Hong Kong Airlines. As noted earlier, the airport is served by full-service network airlines (FSNCs), low-cost carriers (LCCs), dedicated all cargo airlines, and the integrated carriers, for example, FedEx and United Parcel Service (UPS). Hong Kong International Airport is ranked as being one of the world's largest air cargo hubs (Graham & Ison, 2014; Rodrigue, 2020; Sales, 2017).

Founded in 1995, Airport Authority Hong Kong (AAHK) is a statutory body wholly owned by the Government of the Hong Kong Special Administrative Region (HKSAR) and the airport authority is governed by the *Airport Authority Ordinance* (Chapter 483, The Laws of Hong Kong). Guided by the Ordinance and the objective of maintaining Hong Kong's competitiveness as a global and regional aviation hub, the airport authority is responsible for the provision, operation, development, and maintenance of Hong Kong International Airport (HKIA), and the provision of facilities, amenities, or industry at or from any place on the airport island, and other airport-related activities as permitted by the *Airport Authority (Permitted Airport-related Activities) Order* (Cap. 483E) (Airport Authority of Hong Kong, 2021b).

Hong Kong International Airport opened its third runway on 8 July 2022 (Ganesh, 2022; Lee, 2022; Wenzel, 2022). The airport can accommodate International Civil Aviation Organization (ICAO) Code F designated aircraft. The Airbus A380 is an ICAO Code F designated aircraft (Simons, 2014).

Figure 1 presents the annual enplaned passengers handled at Hong Kong International Airport together with the year-on-year change from 2011 to 2020. One passenger enplanement measures the embarkation of a revenue passenger, whether originating, stop-over, connecting or returning (Holloway, 2016). As can be observed in Figure 1, there was considerable growth in Hong Kong International Airport's passenger traffic from 2011 to 2019. Hong Kong International Airport's annual enplaned passengers decreased by 4.13% in 2019 and by 87.63% in 2020 (Figure 1). In 2019, world airline passenger traffic slow downed compared to 2018 (International Air Transport Association, 2020). The global aviation industry has also been adversely affected by the COVID-19 pandemic (Heiets & Xie, 2021; Khan et al., 2022; Lie et al., 2022; Yu & Chen, 2021). The COVID-19 pandemic caused a very substantial reduction in airline passenger traffic (Barczak et al., 2022), and the pandemic had a very disruptive effect on the world air travel market supply and demand chain (Dube et al., 2021). Hong Kong's government implemented a multipronged COVID-19 pandemic response approach, which included border controls, social distancing, quarantine, testing, screening, and surveillance (Wong et al, 2020). Figure 1 shows that there was a very significant decline in Hong Kong International Airport's passenger traffic in 2020, which could be attributed to adverse impact of the Covid-19 pandemic on passenger demand and the related government and airline COVID-19 pandemic response measures.

Fig.1: Hong Kong International Airport annual enplaned passengers and year-on-year change (%): 2011 to 2020. Source: data derived from Hong Kong Civil Aviation Department (2022).

Figure 2 presents the annual aircraft movements at Hong Kong International Airport and the year-on-year change (%) from 2011 to 2020. Figure 2 shows that there was an overall upward trend in the number of aircraft movements during the period 2011 to 2018. However, the airport's annual aircraft movements decreased on a year-on-year basis in 2019 (-1.86%) and 2020 (-61.72%), respectively (Figure 2). The small decrease in 2019 reflected airlines lower aircraft deployment patterns. The significant decrease in 2020 could be attributed to the COVID-19 pandemic impact on air passenger demand as well as the government and airline related response measures to the pandemic. These measures impacted airline aircraft fleet deployment patterns.

Fig.2: Hong Kong International Airport annual aircraft movements and year-on-year change (%): 2011 to 2020. Source: data derived from Hong Kong Civil Aviation Department (2022).

4.2. Hong Kong International Airport Environmental Policy

In October 2011, the Airport Authority of Hong Kong (AAHK) established its first Three-Year Environmental Plan. This plan serves as a principal tool for fulfilling the airport's Corporate Environmental Policy and driving airport-wide environmental improvement. The rolling Plan is updated on an annual basis and covers AAHK's own environmental targets and initiatives as well as efforts with the airport's business partners to reduce the airport's environmental footprint (Airport Authority of Hong Kong, 2014).

In 2012, Hong Kong International Airport made a pledge that the airport would be the world's greenest airport (Airport Authority of Hong Kong, 2015a). A "green airport" is defined as an airport which has a minimal impact on the environment and is an airport that endeavors to become a carbon neutral facility in terms of carbon emissions, with the aim to ultimately produce zero greenhouse gas emissions (González-Ruiz et al., 2017). The concept underlying a "green airport" is for the airport to create a centre of sustainable practices (Sumathi et al., 2018). This goal to be the world's greenest airport serves as a goal and a driver for the airport to continuously improve its environmental performance (Airport Authority of Hong Kong, 2015a).

In 2015, the Airport Authority of Hong Kong defined and implemented a comprehensive corporate environmental policy. In accordance with this policy, the Airport Authority Hong Kong has committed to the long-term sustainable growth of Hong Kong International Airport (HKIA) and is supporting the sustainable development of Hong Kong (Airport Authority of Hong Kong, 2015a). To achieve these goals, the Authority works to minimize the environmental footprint associated with the development and operation of Hong Kong International Airport (HKIA) through:

(1) ensuring compliance with all applicable local and international environmental and related legislation.

(2) optimizing energy and natural resource use efficiencies.

(3) prevention and minimization of both waste and pollution of the land, atmosphere, and surrounding waters of the airport.

(4) implementing an Environmental Management System (EMS) that enables the Authority to manage the company's risks around regulatory compliance, operational resilience, and corporate

reputation, and to create a framework for continual improvement by:

a) identifying and prioritizing key environmental aspects that must include climate change/carbon reduction, air quality, waste management, water usage, ecology & biodiversity, and noise.

b) setting, monitoring, and reporting against clearly defined targets; and

c) assigning appropriate management responsibility for oversight, review, and revision of the Environmental Management System (EMS).

5) providing training to employees and collaborating with business partners to instil a culture of sustainability and extend the most effective measures implemented by the Authority across the whole airport community; and

6) communicating and consulting with employees, relevant airport, government, and community stakeholders to ensure a high level of transparency and that relevant social and economic considerations are considered (Airport Authority of Hong Kong, 2015a).

The Airport Authority is also committed to achieving high standards of environmental performance in pursuit of its pledge to make Hong Kong International Airport the world's greenest airport (Airport Authority of Hong Kong, 2015a).

The Airport Authority of Hong Kong (AA) received its ISO 14001 Environmental Management System (EMS) certification from the British Standards Institution (BSI) on 22 June 2018. This certification formed a key part of Hong Kong International Airport (HKIA) achieving its Greenest Airport Pledge (Airport Authority of Hong Kong, 2018a). The ISO 14001 Environmental Management System (EMS) applies to the environmental aspects of a firm's operations, products, and services that the firm sets and for which it can control and or influence (International Organization for Standardization, 2021; Shehabi, 2016).

On 9 December 2021, Airport Authority of Hong Kong released its target and strategy to achieve Net Zero Carbon by 2050, with a midpoint target of 55% reduction of absolute emissions by 2035 from a 2018 baseline (Airport Authority of Hong Kong, 2021a; Airport World, 2021). Furthermore, the Airport Authority of Hong Kong (AAHK) will work with Technology Working Groups to improve the collaboration between AAHK and business associates (Airport Technology, 2021; International Airport Review, 2021).

The Airport Authority of Hong Kong's sustainability vision is to strengthen the airport's ability to operate and grow profitably in a changing and challenging economic, ecological, technological, and social environment, whilst at the same time developing a robust culture of sustainability throughout the organization (Airport Authority of Hong Kong, 2021b.

4.3. Hong Kong International Airport Water Management System

Hong Kong International Airport (HKIA) uses a "triple water system", that has been designed to improve the efficiency of its three major water sources: freshwater, seawater and treated wastewater. The Airport Authority of Hong's "triple water system" has served the airport since its opening in 1998. A very important benefit of this "triple water system" is that it enables the airport to reduce its freshwater demand by over 50% each year (Airport Authority of Hong Kong, 2022).

Since the airport opened in 1998, Hong Kong International Airport has used seawater for its toilets and air-cooling systems as a standalone component of the airport's triple water system (TWS). The airport's triple water system (TWS) uses potable water for drinking, catering and aircraft washing, and reclaimed water for landscape irrigation. The use of seawater for sanitation and cooling provides substantial cost, energy, and carbon savings over the use of the more traditional "dual water systems", which generally use potable water for these purposes (Airport Authority of Hong Kong, 2013). Potable water is used at the airport in several critical aircraft and airport operations processes, one of which includes aircraft washing (International Civil Aviation Organization, 2020).

As previously noted, at Hong Kong International Airport, seawater is used for toilet flushing and as the cooling medium in the air-conditioning systems of major airport buildings. This significantly reduces the use of freshwater by the airport. Wastewater that is produced from terminal building kitchens, washroom sinks, and aircraft catering and cleaning activities is collected and processed in an on-site wastewater treatment plant. These processed waters are subsequently reused for landscape irrigation (Airport Authority of Hong Kong, 2022).

The Airport Authority of Hong Kong has implemented various measures to manage sewage and storm water discharges at the airport. These measures include:

- Deploying extensive petrol/ oil interceptors in areas where a pollution risk exists, including airport apron areas and airport carparks (Airport Authority of Hong Kong, 2022). The airport apron comprises the individual aircraft stands that interface with the airport terminal building(s) and where aircraft are ground handled in between flights (Budd & Ison, 2017).

- The installation of spill traps at storm water outfalls.
- Discharging storm water away from the potentially sensitive southern sea channel (Hong Kong International Airport, 2022).

The Airport Authority of Hong Kong regularly monitors the airport's impact on the marine environment caused by sewage, storm water discharge and construction activities. The results of this monitoring have indicated that cooling water and storm drain discharges from the airport do not have an adverse impact on local water quality (Airport Authority of Hong Kong, 2022).

4.4 Hong Kong International Airport Annual Water Consumption

Hong Kong International Airport annual water consumption (all sources) and year-on-year change (%) from 2011 to 2020 is presented in Figure 3. Figure 3 shows that the airport's annual water consumption exhibited a downward trend over the period 2011 to 2013, when it decreased from 83,474,000.00 cubic metres in 2019 to 79,697,000.00 cubic metres in 2013. Following this there was a general upward trend from 2014 to 2016, when it increased from 86,444,000.00 cubic metres in 2014 to a high of 92,116,000.00 cubic metres in 2016 (Figure 3). During the latter years of the study, that is, 2017 to 2020, there was an overall downward trend in the airport's annual water consumption. This latter trend was very favorable given the growth in the airport's passenger traffic from 2017 to 2019. The most significant annual increase in water consumption occurred in 2014, at which time it increased by 8.46% on the 2013 levels (Figure 3). There were five years during the study period where the airport's annual water consumption decreased on a year-on-year basis. These annual decreases were recorded in 2012 (-4.43%), 2013 (-0.09%), 2017 (-11.96%), 2018 (-2.74%), and 2020 (-24.18%), respectively (Figure 3). These decreases were favorable as the airport increased its passenger traffic (and aircraft movements) in 2012, 2013, 2017, and 2018, whilst at the same time reducing its municipal water consumption. The significant decrease in 2020 could be attributed to the lower levels of airport operations because of the COVID-19 pandemic and the related government and airline measures that were implemented in response to the pandemic.

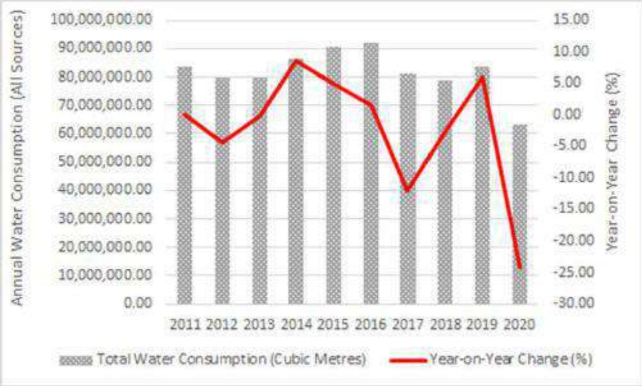

Fig.3: Hong Kong International Airport annual water consumption (all sources) and year-on-year change (%): 2011 to 2020.
Source: data derived from Airport Authority of Hong Kong (2013, 2014, 2015b, 2018b, 2019, 2020, 2021b).

4.5. Hong Kong International Airport Annual Municipal Supplied Water Consumption

Hong Kong International Airport's annual municipal supplied water consumption and the year-on-year change for the period 2011 to 2020 is presented in Figure 4. As can be observed in Figure 4, there were two discernible trends in the airport's annual municipal supplied water consumption during the study period. There was a general upward trend in this metric from 2011 to 2013, at which time it increased from 424,000.0 cubic metres in 2011 to a high of 709,000.0 cubic metres in 2013. There was a pronounced spike in this metric in 2013, when it increased by 61.87% on the 2012 levels. This was the highest single annual increase in this metric during the study period. In 2013, the airport recorded growth in its annual enplaned passengers and aircraft movements, and this translated to additional water requirements. The increase could also be attributed to the airport's stakeholders higher water demand in 2013. Figure 4 shows that there was an overall downward trend in the airport's annual municipal supplied water consumption over the period 2014 to 2020. This is demonstrated by the year-on-year percentage change line graph, which is more negative than positive, that is, all bar value is below the line. In 2017, Hong Kong International Airport's annual municipal supplied water increased by 1.59% on the 2016 levels. This increase was slightly less than the 2016 annual passenger traffic growth rate of 2.97%. Figure 4 shows that there was a significant annual decrease in the airport's municipal supplied water in 2016, at which time it decreased by 38.19% on the 2015 levels. This was also a very favorable outcome as the airport handled higher passenger volumes in 2016, and also handled an increased number of inbound and outbound flights. Figure 4 also shows that the airport's annual municipal supplied water consumption decreased quite significantly in 2014 (-7.47%), 2015 (-7.01%), and 2019 (-7.1%), respectively. Both passenger traffic and aircraft movements increased in 2014, 2015, and 2019 and the airport was able to handle this air traffic growth whilst at the same time lowering its annual municipal supplied water consumption. The lowest annual municipal supplied water consumption was recorded in 2020 (340,000.0 cubic metres) (Figure 4). As previously noted, there was a very pronounced decrease in passenger traffic and aircraft movements in 2020 due to the CORONA-19 virus pandemic and the related government and airline-related response measures.

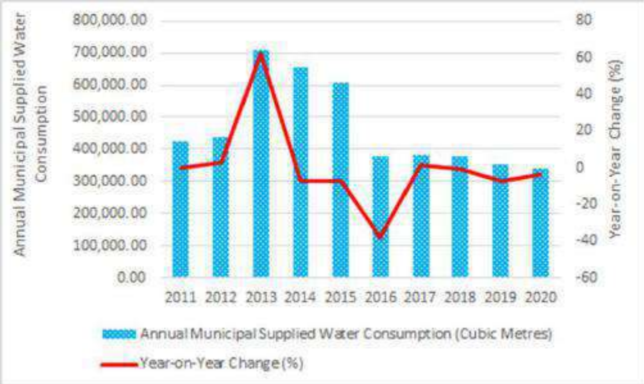

Fig.4: Hong Kong International Airport annual municipal supplied water consumption and year-on-year change (%): 2011 to 2020.
Source: data derived from Airport Authority of Hong Kong (2013, 2014, 2015b, 2018b, 2019, 2020, 2021b).

An important environmental related efficiency metric used by airports is the annual water consumption per enplaned passenger (Baxter, 2022; Graham, 2005) or per workload unit (WLU). One workload (WLU) or traffic unit is equivalent to one passenger, or 100 kilograms of air cargo handled (Doganis, 2005; Graham, 2005; Teodorović & Janić, 2017). Figure 5 presents Hong Kong International Airport's annual water consumption per workload unit (WLU) and the year-on-year change for the period 2011 to 2020. As can be observed in Figure 5, there were four discernible trends in this metric during the study period. In 2012, the annual water consumption per workload unit (WLU) decreased by 2.11% on the 2011 levels (Figure 5). In 2012, the airport's enplaned passengers increased by 5.5%, whilst the annual municipal supplied water consumption increased by 3.3%. Thus, the airport was able to accommodate the higher passenger traffic growth without increasing its water consumption at the same rate of passenger traffic growth. This was a very favorable outcome. Figure 5 shows that there was a pronounced spike in this metric in 2013, at which time it increased by 51.96% on the 2012 levels. This was the second highest annual increase in this metric during the study period. Figure 5 shows that there was a downward trend in this metric during the period 2014 to 2019. This is demonstrated by the year-on-year percentage change line graph, which is more negative than positive, that is, all values are below the line. The annual municipal supplied water consumption per workload (WLU) declined significantly in 2014 (-12.79%), 2015 (-14.09%), and 2016 (-40.29%), respectively (Figure 5). In each of these years, the airport was able to accommodate and process higher levels of passenger traffic, whilst at the same time reducing the amount of municipal supplied water consumed. This was another very favorable outcome for the airport and demonstrated a high level of water consumption efficiency. As can be observed in Figure 5, there was a very pronounced spike in this metric in 2020, when it increased by 678.94% on the 2019 levels. In 2020, the airport reduced its annual municipal supplied water consumption by 3.68%. However, in 2020, the airport's annual passenger throughput decreased by 87.63%, and, as a result, there were fewer passengers to spread the water consumption over. This translated into the significant spike in this metric in 2020.

Fig.5: Hong Kong International Airport annual municipal supplied water consumption per workload unit (WLU) and year-on-year change (%): 2011 to 2020.
Source: data derived from Airport Authority of Hong Kong (2013, 2014, 2015b, 2018b, 2019, 2020, 2021b).

Hong Kong International Airport's annual municipal supplied water consumption as a portion of the airport's total annual water consumption and the year-on-year change from 2011 to 2020 is presented in Figure 6. As can be observed in Figure 6, Hong Kong International Airport's annual municipal supplied water as a portion of the airport's total annual water consumption has oscillated throughout the study period. From 2011 to 2013, there was a general upward trend in this metric, when it increased from 0.51% in 2011 to a high of 0.89% in 2013 (Figure 6).

The most significant annual increase in this metric was recorded during this period. In 2013, the metric increased by 61.81% on the 2012 levels. As previously noted, Hong Kong International Airport's annual municipal supplied water increased very significantly in 2013 (+61.87%), and this in turn resulted in municipality supplied waters accounting for a greater share of total water consumption in 2013. Figure 6 shows that this metric decreased quite significantly in 2014 (-14.60%), 2015 (-11.84%), and 2016 (-38.80%), respectively. These decreases could be attributed to the airport's lower annual municipality supplied water consumption in these respective years. Figure 6 also shows that there were two quite significant annual increases in this metric during the study period. These increases occurred in 2017 (+14.63%), and 2020 (+28.57%) (Figure 6). The increase in 2017 could be attributed to the higher municipal supplied water consumption at the airport during 2017. In 2020, the annual municipal supplied water consumption decreased by 3.68%, whilst the annual seawater consumption, a key water source, decreased by 24.27% in 2020. As a result, municipal supplied water consumption as a share of total water consumption increased in both 2017 and 2020, reflecting the differing water sources consumption patterns in 2020.

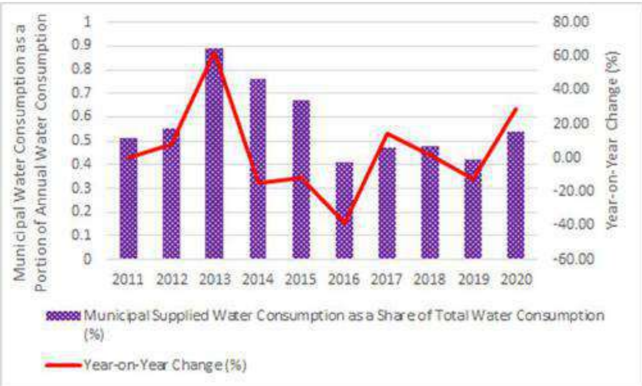

Fig.6: Municipal supplied water as a share of total annual water consumption and year-on-year change (%): 2011 to 2020.
Source: data derived from Airport Authority of Hong Kong (2013, 2014, 2015b, 2018b, 2019, 2020, 2021b).

4.6. Hong Kong International Airport Annual Seawater Consumption

Hong Kong International Airport's annual seawater consumption and the year-on-year change from 2011 to 2020 is depicted in Figure 7. Figure 7 shows that there were three discernible trends in the airport's annual seawater consumption. There was a general downward trend during the early years of the study, that is, 2011 to 2014, at which time the airport's annual seawater consumption decreased from 83,050,000 cubic metres in 2011 to 78,988,000 cubic metres in 2013. In these early years of the study period, the most significant annual decrease was recorded in 2012, when it decreased by 4.47% on the 2011 levels, reflecting a lower usage requirement in 2012. From 2014 to 2016, there was a general upward trend in the airport's annual seawater consumption, with the two most significant annual increases being recorded in 2014 (+8.6%), and 2015 (+5.11%), respectively (Figure 7). Hong Kong International Airport recorded strong growth in its annual passenger traffic (and aircraft movements) in both 2014 and 2015, and this growth in passenger traffic and aircraft movements would have required greater amounts of water to sustain the airport's operations. From 2017 to 2020, there was an overall downward trend in the airport's annual seawater consumption, when it decreased from a high of 91,739,000 cubic metres in 2016 to a low of 63,069,000 cubic metres in 2020. Figure 7 shows that there was one exception to this downward trend, when the airport's annual seawater consumption increased by 6.11% in 2019, reflecting higher consumption patterns. In 2020, the airport's annual seawater consumption decreased by 24.27% on the 2019 levels, reflecting the lower passenger volumes and aircraft movements in 2020 handled at the airport due to the CORONA-19 virus pandemic.

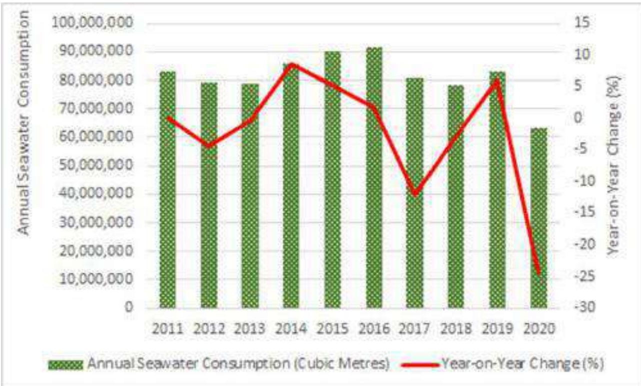

Fig.7: Hong Kong International Airport annual seawater consumption and year-on-year change (%): 2011 to 2020.
Source: data derived from Airport Authority of Hong Kong (2013, 2014, 2015b, 2018b, 2019, 2020, 2021b).

Hong Kong International Airport's annual seawater consumption as a portion of the airport's total annual water consumption and the year-on-year change from 2011 to 2020 is presented in Figure 8. Figure 8 shows that this metric has fluctuated over the study period reflecting differing consumption patterns. During the study period, there were four years where the airport's annual seawater consumption as a share of total water consumption increased slightly on a year-on-year basis. These annual increases occurred in 2014 (+0.13%), 2015 (+0.09%), 2016 (+0.26%), and 2019 (+0.06%), respectively (Figure 8). In 2014, the airport's annual municipal supplied water consumption decreased on a year-on-year basis whereas the airport's seawater consumption increased in the same year, and thus, this resulted in the seawater's higher share of total water consumption in 2014. A similar situation occurred in 2015, when once again the airport's municipal supplied water consumption declined on a year-on-year basis, whilst at the same time the annual seawater consumption in that year, and once again, this resulted in the airport's seawater consumption accounting for a higher share of the airport's total annual water consumption in 2015. The same trend occurred in 2016, when once again the airport's municipal supplied water consumption decreased on an annualized basis, whilst the airport's seawater consumption increased on an annualized basis. Hence, in 2016, the airport's annual seawater consumption once again accounted for the higher share of the airport's total annual water consumption in that year. In 2019, there was a significant decrease in the airport's municipal supplied water and a 6.11% increase in the annual seawater consumption. This translated into the growth in the seawater consumption as a share of the airport's total annual water consumption in 2019. Figure 8 shows that there were five years in the study period where the airport's annual seawater consumption as a share of total water consumption decreased on a year-on-year basis. These annual decreases occurred in 2012 (-0.04%), 2013 (-0.34%), 2017 (-0.06%), 2018 (-0.01%), and 2020 (-0.12%), respectively (Figure 8). In 2012 and 2013, the airport's annual seawater consumption declined on a year-on-year basis whereas the airport's annual municipal supplied water increased in both years. This resulted in an increase in the municipal supplied share of total water consumption and a decrease in the seawaters share of total water consumption in both years. A similar trend occurred in 2017, at which time the airport's aannual municipal supplied water consumption increased on a year-on-year basis, whilst at the same time the annual seawater consumption decreased by -12.02%. Consequently, the seawater share of the airport's total annual water consumption declined whilst the municipal supplied water consumption as a share of total water increased in 2017. In 2018, the airport consumed increased volumes of municipal supplied water but consumed less seawater. Once again, this meant that there was a decrease in the seawater consumption as a share of total water consumption in 2018. In 2020, the airport's annual municipal supplied water and seawater consumption both decreased on an annualized basis due to the impact of the CORONA-19 virus pandemic and the related government and airline related pandemic response measures. However, in 2020, the decrease in the airport's annual seawater consumption was lower than the decrease in the airport's municipal supplied water consumption, and hence, the seawater consumption as a share of total water consumption was influenced by the differences in the two water sources annual consumption pattern.

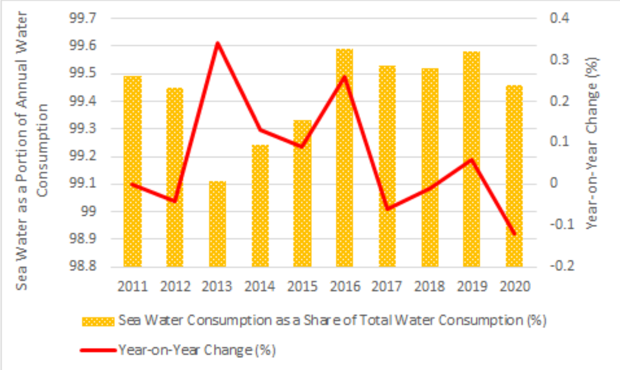

Fig.8: Sea water as a share of total annual water consumption and year-on-year change (%): 2011 to 2020. Source: data derived from Airport Authority of Hong Kong (2013, 2014, 2015b, 2018b, 2019, 2020, 2021b).

4.7. Hong Kong International Airport Annual Recycled/Re-Used Water Consumption

As noted earlier, Hong Kong International Airport re-uses and recycles wastewaters, which once processed the re-processed waters are used for irrigation purposes. It is important to note that the re-use of water produces substantial environmental benefits, arising from reductions in water diversions, and reductions in the impacts of wastewater discharges on environmental water quality (Anderson, 2003). The re-use of wastewater also prevents environmental pollution (Nair, 2008; Ofori et al., 2021). Hong Kong International Airport's annual recycled/re-used waters and the year-on-year change from 2011 to 2020 is presented in Figure 9. As can be observed in Figure 9, Hong Kong International Airport's annual recycled/re-used waters have oscillated over the study period. Figure 9 shows that there were two quite pronounced annual decreases in this metric, with these decreases occurring in 2013 (-26.66%) and 2018 (-16.41%), respectively. These decreases were due to smaller volumes of water that were available to be recycled/re-used in both 2013 and 2016. Figure 9 shows that were was a spike in this metric in 2014, at which time the annual waters recycled/re-used increased by 16.23% on the 2013 levels. There were four smaller annual increases in this metric during the study period, with these smaller annual increases occurring in 2012 (+5.52%), 2014 (+3.24%), 2016 (+8.1%), and 2019

(+6.13%), respectively (Figure 9). This was a very favorable outcome for the airport and showed that it was able to make very good use of its waters in these respective years. Figure 9 also shows that the annual recycled/re-used waters decreased in 2020 by 2.89% in 2020, reflecting the lower water requirements at the airport due to the CORONA-19 pandemic government and airline response measures. During the study period, the smallest annual volume of recycled/re-used waters was recorded in 2013, at which time the airport recycled/reused 154,000 cubic metres of water (Figure 9). The highest annual volume of recycled/re-used waters was recorded in 2012, at which time the airport recycled/reused 210,000 cubic metres of water (Figure 9).

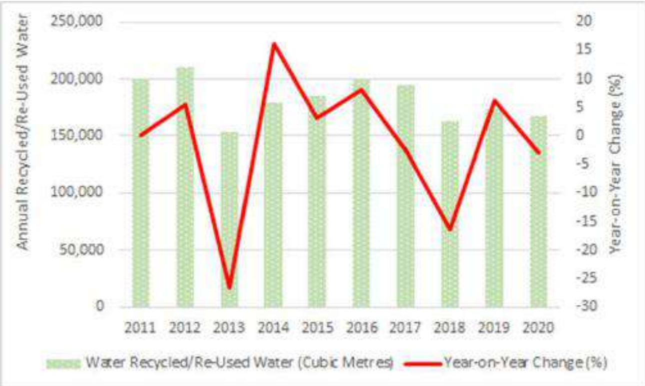

Fig.9: Hong Kong International Airport annual recycled/re-used waters and year-on-year change (%): 2011 to 2020.

Source: data derived from Airport Authority of Hong Kong (2013, 2014, 2015b, 2018b, 2019, 2020, 2021b.

4.8. Hong Kong International Airport Annual Discharged Waters

Prior to examining Hong Kong International Airport's annual discharged wastewaters, it is important to note that very significant volumes of wastewater can be produced at airports (Baxter et al., 2018a; Pitt et al., 2002). Allenby and Park (2013, p. 462) have observed that wastewater is "water that carries wastes from homes, businesses, and industries and usually contains dissolved solids and/or suspended solids". Hong Kong International Airport's annual discharged wastewaters and the year-on-year change for the period 2011 to 2020 is presented in Figure 10. Figure 10 shows that the airport's annual discharged wastewaters fluctuated over the study period. There was a very pronounced spike in the airport's annual discharged wastewaters in 2013, at which time they increased by 143.42% on the 2012 levels. In 2013, the airport discharged a total of 555,000 cubic metres of wastewaters. This was the highest annual discharge of wastewaters by the airport during the study period. The smallest annual volume of discharged waters was recorded in 2020, at which time the airport discharged 172,000 cubic metres of wastewaters (Figure 10). Figure 10 also shows that there were three years during the study period where smaller annual increases were recorded in the annual wastewaters discharged by the airport. These increases were recorded in 2012 (+1.33%), 2017 (+6.21%), and 2018 (+15.42%) and reflected a greater requirement by the airport to discharge waters in these respective years (Figure 10). Figure 10 also reveals that there were five years during the study period where the annual volume of wastewaters discharged decreased on a year-on-year basis. These annual decreases occurred in 2014 (-14.05%), 2015 (-10.9%), 2016 (-58.35%), 2019 (-17.05%), and 2020 (-4.44%), respectively (Figure 10). These annual decreases are another very favorable trend and in these years the airport was able to mitigate the potential level of environmental harm from its annual wastewater discharges. An airport's annual wastewaters can fluctuate (Baxter, 2021; Baxter et al., 2018a, 2018b) and Hong Kong International Airport's annual wastewaters have displayed such a trend.

The volume of wastewater discharge at Hong Kong International Airport is estimated by subtracting the volume of wastewater recycled from the airport's municipal water consumption. Under the *Water Pollution Control Ordinance* (Chapter 358, The Laws of Hong Kong), the Airport Authority of Hong Kong AAHK holds a number of licenses which require the monitoring of water quality using the following parameters: flow rate (m^3/day), total residue chlorine, amines, temperature, antifoulant, suspended solids, chemical oxygen demand, oil and grease, surfactants (total), biochemical oxygen demand, total phosphorus and formaldehyde (Airport Authority of Hong Kong, 2021b).

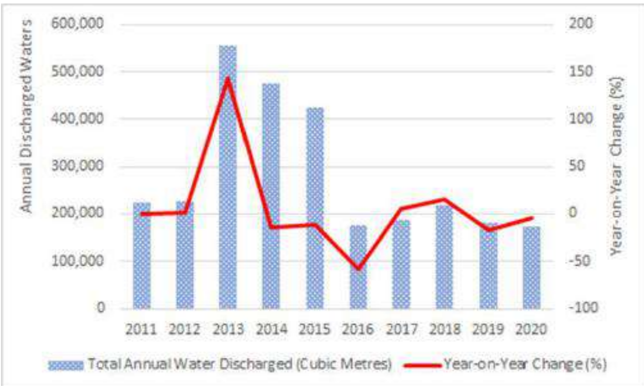

Fig.10: Hong Kong International Airport annual discharged wastewaters and year-on-year change (%): 2011 to 2020.

Source: data derived from Airport Authority of Hong Kong (2013, 2014, 2015b, 2018b, 2019, 2020, 2021b).

4.9. Dry Aircraft washing at Hong Kong International Airport

In June 2019, the Airport Authority of Hong Kong granted approval for Hong Kong Aircraft Engineering Company Limited (HAECO) to perform the dry washing of aircraft at the airport (International Civil Aviation Organization, 2020). Aircraft dry washing involves applying cleaning agents to the aircraft surface, then removing with mops or wipes. This practice uses considerably less water and can be carried out in any location (Hayward, 2022). The cleaning product used in the dry washing of aircraft is more effective at removing insects, oil stains and other dirt from the aircraft's exterior. Following washing, a fine protective film is formed on the aircraft enabling it to retain a longer shine. Importantly, the aircraft dry wash technique uses 90% less water than conventional wet washing methods that use highly pressurized water, while the biodegradable, non-toxic cleaning product produces fewer chemical effluents (Airport Authority of Hong Kong, 2020b).

Traditionally, aircraft washing at Hong Kong International Airport was allowed only in 10 designated aircraft parking bays that were equipped with drainage to collect effluent for treatment. Prior to approving the dry washing of aircraft for a further 29 predesignated aircraft parking bays, which reduces the distance of aircraft towing by relaxing the requirements to use parking bays for cleaning, the airport authority reviewed the physical and chemical properties of the products, assessed the environmental risks, and provided advice on the Dry Wash Procedures. The benefits of dry wash are significant in various aspects. Compared with the traditional wet washing of aircraft, HAECO anticipates an aircraft dry wash to use 90% less water. This new practice saves more than 860,000 liters of water a year and produces less effluent. An additional environmental related benefit is that by reducing the aircraft towing requirement, this helps reduce the traffic on the apron and fuel consumption by aircraft and ground services equipment (GSE), hence this reduces the airport-wide greenhouse gas emissions (International Civil Aviation Organization, 2020). In 2020, the Airport Authority of Hong Kong approved another aircraft maintenance service provider, China Aircraft Services Limited (CASL), to conduct aircraft dry wash on the maintenance apron at the airport (Airport Authority of Hong Kong, 2020a; International Civil Aviation Organization, 2020).

4.10. Hong Kong International Airport Water Saving Efficiency Measures in the Midfield Concourse (MFC)

Hong Kong International Airport has installed a range of water saving efficiency measures in its new Midfield Concourse (MFC). A sustainable water strategy has been adopted for the MFC which covers demand reduction, grey water recycling, and condensate water harvesting. The airport's goal is to reduce potable water consumption by 30% compared to typical Hong Kong consumption. The water demand reduction is principally supported using sea water for toilet flushing and other water-conserving sanitary fittings. Treated grey water and condensate water is to be reused in the cooling system of the MFC to further reduce potable water consumption. The airport's Midfield Concourse (MFC) water strategy aims to eliminate water leakage, reduce water consumption, use non-potable water for the flushing of toilets, and harvest and recycle water where possible (Airport Authority of Hong Kong, 2015). In addition, the airport is using condensate water from air-conditioning systems as well as reclaimed water to cool the chiller systems in the concourse, and these measures will thereby reduce the use of potable water (Airport Authority of Hong Kong, 2014).

V. CONCLUSION

Utilizing an in-depth longitudinal case study research design, this study has examined Hong Kong International Airport's water management and the annual trends in the airport's water consumption for the study period of 2011 to 2020. The qualitative data used in the study was examined by document analysis.

The case study revealed that the two principal sources of water used by the airport are municipal supplied water and seawater. The airport also recycles water for use in the irrigation of the airport's grounds. The airport uses a "triple water system", that has been designed to improve the efficiency of its three major water sources: freshwater, seawater and treated wastewater.

The airport's total annual water consumption decreased from 83,474,000.0 cubic metres in 2011 to a low of 63,409,000.0 cubic metres in 2020. The highest annual water consumption was recorded in 2016, when the airport consumed 92,116,000.00 cubic metres of water.

The case study found that the airport's use of municipal supplied water displayed an upward trend from 2011 to 2013 and an overall downward trend from 2014 to 2020. This latter trend was very favorable as the airport's annual enplaned passengers and aircraft movements grew very strongly over the study period. The municipal supplied water consumption per enplaned passenger or per workload unit (WLU) largely displayed a general downward trend. The lowest annual municipal supplied water consumption per enplaned passenger or per workload unit (WLU) was recorded in 2019 (4.94 litres/WLU), whilst the highest was recorded in 2020 (38.48 litres/WLU). The airport was adversely impacted by the CORONA-19 virus pandemic and the related government response measures, and thus, there were fewer passengers handled at the airport and this led to the very large increase in 2020.

The airport's annual seawater consumption oscillated over the study period, reflecting differing consumption patterns. The lowest annual seawater consumption was recorded in 2020, at which time the airport consumed 63,069,000 cubic metres of seawater. The highest annual seawater consumption was recorded in 2016, at which time the airport consumed 91,739,000 cubic metres of seawater. Sea waters are the primary water source used by the airport, averaging around 99.43% of the airport's total annual water consumption during the study period.

The case study also found that Hong Kong International Airport's annual recycled/re-used water consumption fluctuated over the study period. The lowest annual recycled/reused waters consumption was recorded in 2013, at which time the airport consumed 154,000 cubic metres of recycled/re-used waters. The highest annual recycled/re-used water consumption was recorded in 2012, at which time the airport consumed 210,000 cubic metres of recycled/re-used waters.

Hong Kong International Airport's annual discharged wastewaters also fluctuated over the study period. The lowest annual level of discharged waters was recorded in 2020, at which time the airport consumed 172,000 cubic metres of discharged waters. The highest annual release of discharged waters was recorded in 2013, at which time 555,000 cubic metres of wastewaters were discharged from the airport.

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Influence of different levels of nitrogen fertilizers on some sunflower cultivars quality: A Review

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***Abstract*— *The effect of NF on sunflower cultivars of Flamy (FL) and Manon (MA) were tested during planting at three NF of 175, 200 and 250 kg.N.ha^{-1}. The experiments were conducted in a factorial experiment under complete randomized design with three replications. The results showed that the MA cultivar was significantly better than the FL in all studied conditions. The soil properties SBD, TSP, RL, RDFW, PDFW, SD, Sl, PVI and GY were; 1.29mg.cm^{-3} ,1.30 mg.cm^{-3},1.33 mg.cm^{-3}, 51.32%, 50.94%, 49.81% ,24.115 cm , 161.419 g, 184.624 g, 38.01 mm, 22.58mm , 80.39 cm and 5.311 t.ha^{-1}, respectively. The NF of 250 kgN.ha^{-1} was significantly superior to the levels of 175 and 200 kg N.ha^{-1} in all studied conditions.*

***Keywords*— sunflower, nitrogen fertilizers(NF), Flamy(FL), Manon(MA).drip irrigation system.**

I. INTRODUCTION

Sunflower crop (*Helianthus annuus L*.), is most important and strategic oilseed crops of the compound family Asteraceae, its bears difficult climatic conditions when drought or high temperatures, so its cultivation is successful in various climatic conditions, Iraq suffers from a large deficit in the production of vegetable oils. Therefore, attention must be paid to the cultivation of oil crops, the most important of which is the sunflower crop due to the high percentage of oil in its seeds, as it reaches more than 45%, in addition, sunflower oil is characterized by the quality of its chemical and natural properties, also it contained a high protein percentage and was considered an important source in the preparation of animal feeds (Alsharifi.,2009; Iqbal et al.,2021). The Iraqi Ministry of Agriculture sought after 2021, because of the food crisis that the world is going through, it launched the Food Security Initiative and took the administrative and financial responsibility to encourage farmers to plant important agricultural crops that support national food security the most important crops are wheat, barley, rice and corn, also focused on cultivation of oil crops to provide the table oil that the Iraqi family needs as a main source in cooking food. (Alsharifi et al.,2021a; Alaamer et al.,2021; Alsharifi et al.,2022).

The study of (Alsharifi and Ameen 2018; Shtewy et al.,2020a; Al-Jezaaria et al.,2021, showed that planting any crop requires creating a suitable environment for germination by choosing the appropriate machine to complete all agricultural operations from tillage, smoothing, levelling and dividing the field to increase the germination ratio, ease carrying out of crop service operations, throughout the plant growth period until full maturity, important agricultural processes affecting the crop out put, include methods adding of fertilizers formulations and organics acids , as well as planting distances between plants.(Al-Zubaidi, and Al-Awsi 2017). Scientifics basis confirms is the soil analysis before adding fertilizers to know the plant's extent need for the amount and fertilizer type added on the soil's loss basis of it, unless it is organic or chemical, determining the type and fertilizers quantity added to the soil, reflects the chemical and physical properties improvement of the soil and an increase in the plants output.(Alaamer et al.,2021b;

Alsharifi et al.,2022). Adding chemical fertilizers and organic acids in increasing quantities to the soil by means of drip irrigation system or immersion irrigation, after their decomposition in soil , they are absorbed by the plants roots and this is reflected in the balance state within the plant tissues and thus an increase in the plant growth characteristics, the reason for this is due to the improvement of the soil qualities, which were suffering from an acute nutrients shortage for the plant (Al-Mughair .2019; and Ahmed and Al-Tamimi .2020). The study of (Alsharifi et al.,2020a; Shtewy et al.,2020b) showed that Increasing the amount of chemical fertilizers added to the soil may lead to a deterioration in the soil condition resulting from hardening it and preventing the root spread. Therefore, it is preferable to use organic fertilizers, which in turn lead to the fragmentation of the soil (soil fragility), and the improvement of its physical and chemical properties, and the spread of roots abundantly and in great depths, and this is reflected in the improvement of all growth characteristics and yield (Abdullah, 2008; Awais et al.,2015). The aim of the article was to evaluate some plant sunflower traits for two Flamy (FL) and Manon (MA) cultivars at different levels of nitrogen fertilizer.

II. MATERIALS AND METHODS

This study was conducted in 2021, to evaluate some plant sunflower traits under effect nitrogen fertilizers. The experiments were done at three levels of nitrogen fertilizers (NF) of 175, 200 and 250 kgN.ha^{-1}, and two Flamy (FL) and Manon (MA) cultivars . In this study the MF390 tractor use with sweep plow on depth of 20-24 cm to soil stir and prepare a pot suitable for germination, chemical fertilizers were added (DAB type), at a rate of 400 kg.ha^{-1}, then the field was divided according to the planting distances planned in the experiment 25 * 75 (Fig .1), after which the seeds were planted using the planting machine (Blanter type). drip irrigation system was used in this experiment, according to the method used by (Alsharifi et al.,2021b). Nitrogen was added at the first batch after 4 weeks from germination and the second batch added during flowering stage and phosphorous at one level 50 kg N.ha^{-1}, during growth season, Al-Sharifi et al.,2020. The root length (RL), root dry and fresh weight (RDFW), plant dry and fresh weight (PDFW), stem diameter (SD), seed length (SL) PVI, and grain yield (GY), were calculated for each running test.

Calculation of the injection rate, for fertilizer an area of one hectare

$$. Q_F = \frac{F_{R \times A}}{T_{I \times T_{F \times F_C}}}$$

Where ;Q_F ;fertilizer injection rate L.ha^{-1},F_R ;fertilizer rate kg.ha^{-1} , A; The area to be fertilized ha,T_I Irrigation time hour ,T_F ;ratio of fertilizing time to irrigation time ,F_C ; Fertilizer concentration kg.L^{-1}

$$V = \frac{F_{R \times A}}{F_C}$$

Where**; V,** Tank capacity

$$X = F_{C \times R}$$

Where; X the amount of fertilizer added to the tank of the drip irrigation system.

$$Q = \frac{F_{pm}}{C \times 10^6} = Q_{F \times F_C}$$

Or

$$Q = \frac{V}{C} \times \frac{Q}{F_C} \times \frac{EPP}{10^6}$$

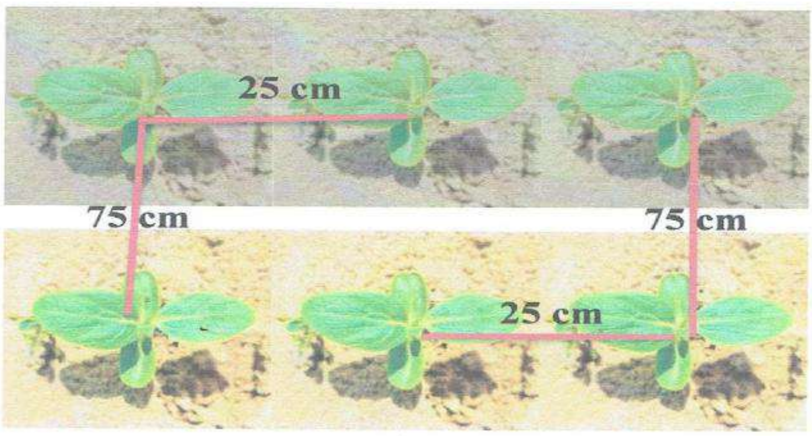

Fig .1. Field cultivation distances for sunflower crop

2.1. Soil texture:

All soil properties (soil density and porosity) were calculated, during the plant growing season, as random samples were taken for several locations in the experimental field after a month of germination (1Mon), two months of germination (2Mon) , the season end (SE) ,according to the methods used by (Al-Jezaaria et al.,2021; Alaamer et al.,2020) the equations below were used to calculate it. (Hu et al.,2012; Hamzah and Alsharifi,2020)..

$$W = \frac{W_w}{W_s} \times 100 \quad (1)$$

Where: W is soil humidity ratio (%), W_w is mass wet soil(kg), W_s is mass dry soil.**(**kg**)**

$$P_b = \frac{M_S}{V_T} \quad (2)$$

Where: P_b: (mg. m^{-3}), M_S: (mg), V_T: total volume (m^3).

$$T_{SP} = \left(1 - \frac{P_b}{P_S}\right) \times 100 \quad (3)$$

Where: T_{SP}: (%), P_b: (mg.m^{-3}), P_S : partial density (2.65 mg.m^{-3}). [26]

Table 1. Chemical and physical analysis of soil particles

Depth	Texture %			
0-25 (cm)	Clay	Silt	Sand	
	45	30	25	Silt Clay loam
	Soil physical properties			
	Pb (Mg m^{-3})	TSP (%)	SPR (Kpa)	
	1.36	48.67	1766.423	
	1.38	47.92	1798.090	
	1.41	46.79	1891.315	
VA	1.38	47.79	1818.609	
0-25	Soil chemical properties			
	E.C (ds\cm^3)	HP		
	1.61	6.33		
	Soluble cation meq\I			
	Na	K	Ca+Mg	
	11.14	13.24	56.20	
	O.C (%)	CEC (Meq\100g)	CaCo3 (%)	O.M (%)
	0.58	32.01	4	0.64

2.2. The Crop and Its Attributes

2.2.1. Root Length (RL)

It was calculated according to (Shtewy and Alsharifi .,2020c)

2.2.2 RDFW

2.2.3.PDFW

2.2.4.Stem Diameter (cm): Calculated using the (Vernier machine). (Bajehbaj et al.,2009;Shtewy et al.,2020b)

2.2.5..Seed length (mm): Calculated using (Vernier machine)

2.2.6**. Plant vigor index (PVI)**

Was calculated according to (Ghali et al.,2020 Shtewy et al., 2020a)

$$P_{VI} = \frac{P_L \times G_P}{100}$$

Where ; PVI ;plant vigor index cm, P_L ;plant length cm, G_P;Germination ratio.

. **2.2.7.Grains yield;**

The grains yield was calculated (Alsharifi 2009, Alsharifi et al,2021c)

$$G_{Y=GP\times PD}$$

Where ; G_Y;grain yield(t.ha^{-1}),GP; grain rate per plant (kg) , PD; plant density.ha^{-1}.

Fig 2. Field experiment

The obtained results were analysed in the field according to the method approved by .(Oehlent ,2010)..

III. RESULTS AND DISCUSSION

Table (2) shows treatment NF of 175kg N.ha^{-1} reduced the SBD values, after a one month, two months, growing season end, and the results were 1.28,1.29 and 1.32 Mg. m^{-3}, offset by an increased in the TSP, were results,51.69, 51.32 and 50.18% respectively, after a 1Mon, 2Mon and GSE, with the same situation for the NF of 200 and 250 kg N.ha^{-1}), (Hu et al.,2012 , Alsharifi et al.,2021a). The sunflower cultivar (MA), done the best SBD of 1.29,1.30 and1.33Mg.m^{-3}, and the excess in ratios TSP of 51.32, 50.94and 49.81%, with the same situation for the sunflower cultivar (FL).

Table (3) showed that the NF had a significant effect on plant traits and roots, RL, RDFW and PDFW, as the NF of 250 kgN.ha^{-1} exceeded it and gave the highest average of 25.297 cm ,163.109 g and 182.948 g respectively , while the NF of 175kg N.ha^{-1} treatment gave the lowest average of 21.746 cm, 153.737 g and 178.810 g. Perhaps the reason for this is that increasing the fertilizer amount added to the soil improves the plant growth properties by increasing the roots effectiveness and excess their depth in the soil., the results agreed with what was reached by (Shtewy et al.,2020c; Al-Zubaidi and Al-Awsi.,2017),. All plant traits and roots excessed with MA cultivar and scored the higher results 24.115 cm,161.419g and 184.624 g, as compared with FL cultivar which gave the lower results22.471cm, 156.892 g and 177.251g, respectively, (Al-Mughair, 2019;Alaamer et al.,2021a). The interaction among MA cultivar and the NF of 250kgN.ha^{-1} was the best (26.428 cm, 1165.728 g and 1186.775 g). The levels of the plant traits and roots at various conditions are shown in Figure 3 for both Cu and NF.

Table (4) showed that the NF had a significant effect on plant traits and productivity , SD, SL, PVI and GY, the NF of 250 kgN.ha^{-1} exceeded it and gave the highest average of 40.54 mm, 24.88 mm, 80.37 cm and 5.415 t.ha^{-1} respectively , while the NF of 175kg N.ha^{-1} treatment gave the lowest average of 32.81 mm,18.66 mm, 78.53 cm and 4.195 t.ha^{-1}respectively for SD, SL, PVI and GY. Perhaps this is due to the increased plant growth effectiveness and thus increase its productivity, when the fertilizer amount added to the soil increased., the results agreed with what was reached by (Abdullah ,2008; Ahmedand Al-Tameemi, 2020). All plant traits and roots excessed with MA cultivar and scored the higher results 38.01 mm, 22.58 mm, 80.39 cm and 5.311t.ha^{-1} , as compared with FL cultivar which gave the lower results34.29 mm, 20.90 mm, 78.34 cm and 4.105 t.ha^{-1}, respectively, reason for this is the MA cultivar nature and its tolerance to harsh growing conditions such as drought and high temperatures, its ability to achieve the best results of this study. (Awais et al.,2015; Alsharifi et al.,2022). The interaction among MA cultivar and the NF of 250kg N.ha^{-1} was the best 42.86mm, 26.07mm, 81.09 cm and 6.009t.ha^{-1} respectively. The levels of the plant traits and productivity at various conditions are shown in Figure 4 for both Cu and NF.

Table 2. influence of NF levels and Cu on soil properties

Cultivars (Cu)	NF	Soil bulk density			Total of soil porosity		
		1Mon	2Mon	SE	1Mon	2Mon	SE
MA	175	1.26	1.28	1.30	52.45	51.69	50.94
	200	1.30	1.31	1.34	50.94	50.56	49.43
	250	1.31	1.32	1.36	50.56	50.18	48.67
FL	175	1.29	1.30	1.33	51.32	50.94	49.81
	200	1.31	1.32	1.35	50.56	50.18	49.05
	250	1.34	1.36	1.40	49.43	48.67	47.16
Cu	MA	1.29	1.30	1.33	51.32	50.94	49.81
	FL	1.31	1.33	1.36	50.56	49.81	48.67
NF	175	1.28	1.29	1.32	51.69	51.32	50.18
	200	1.30	1.32	1.35	50.94	50.18	49.05
	250	1.33	1.34	1.38	49.81	49.43	47.92
LSD=0.05	MA	0.02	0.03	0.04	0.148	0.154	0.163
	FL	0.04	0.05	0.06	0.155	0.163	0.176
	MA*FL	0.05	0.06	0.07	0.203	0.212	0.255

Table 3. Impact of NF and Cu on plant traits and roots

Cultivars (Cu)	NF	RL cm	RDFW g	PDFW g
MA	175	22.411	156.403	182.415
	200	23.505	162.126	184.682
	250	26.428	165.728	186.775
FL	175	21.081	151.072	175.206
	200	22.175	159.115	177.427
	250	24.166	160.491	179.122
Cu	MA	24.115	161.419	184.624
	FL	22.471	156.892	177.251
NF	175	21.746	153.737	178.810
	200	22.840	160.621	181.054
	250	25.297	163.109	182.948
LSD=0.05	MA	1.314	1.425	2.504
	FL	1.508	1.609	2.624
	MA*FL	1.817	1.913	3.086

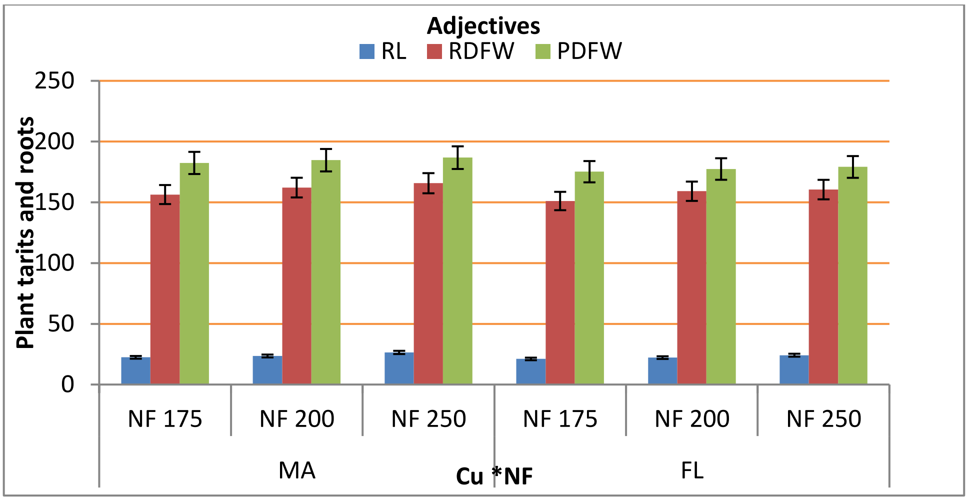

Fig 3. Impact of NF and Cu on plant traits and roots

Table 4. Impact of NF and Cu on plant traits and productivity

Cultivars (Cu)	NF	SD mm	SL mm	PVI cm	GY $t.ha^{-1}$
MA	175	34.62	19.18	79.92	4.903
	200	36.55	22.51	80.16	5.021
	250	42.86	26.07	81.09	6.009
FL	175	31.01	18.13	77.13	3.488
	200	33.65	20.87	78.24	4.008
	250	38.22	23.69	79.65	4.819
Cu	MA	38.01	22.58	80.39	5.311
	FL	34.29	20.90	78.34	4.105
NF	175	32.81	18.66	78.53	4.195
	200	35.10	21.69	79.20	4.514
	250	40.54	24.88	80.37	5.415
LSD=0.05	MA	1.521	1.526	1.326	0.142
	FL	1.509	1.719	1.551	0.214
	MA*FL	2.133	2.113	2.675	0.423

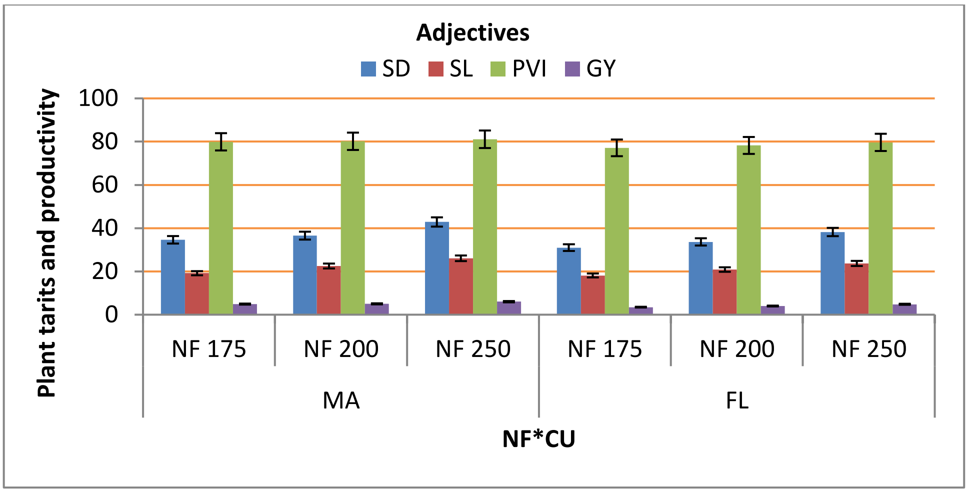

Fig. 4. Impact of NF and Cu on plant traits and productivity

IV. CONCLUSIONS

The sunflower cultivar of MA, was exceedingly significantly than sunflower cultivar of FL. Also the level NF of 250 kg N.ha^{-1},was superior significantly than two levels other NF 175kg N.ha^{-1}and 200 kg.N.ha^{-1}), in all studied conditions. were higher results when interaction between MA cultivar and NF Of 250 kg N.ha^{-1}) , and in all studied conditions.

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Soil moisture variations of wetland at different altitudes in desert regions, China

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Abstract— *Desert wetland is a special ecological environment as water is fundamental to maintaining existence. The soil water of sandy wetland links surficial water and groundwater. This study Takes Habahu National Wetland Nature Reserve as the research area and the water movement characteristics of soil profile as the research purpose. According to the altitude and topographic characteristics, the main focus is monitoring soil moisture in different layers at specific sites. The results show that: (1) In the core area of sandy wetland, the water movement of the soil profile is slow with lower infiltration and evaporation; (2) On the slopes around the wetland, the surface water is recharged by rapid leakage along with the underlying impermeable layer after precipitation due to the sand coverage; (3) In the desert hinterland outside the wetland, the water rapidly infiltrates after precipitation and flows into different groundwater systems along the structural characteristics of the underlying stratum to supplement the nearby wetlands. Based on these characteristics, we put forward the unique viewpoints of environmental protection in sandy wetlands: (1) focus on the protection of surface organisms in the core area of sandy wetland to reduce evaporation and improvethe soil water-holding capacity; (2) Appropriate engineering and biological measures should be applied to the slope outside the core area to reduce surface evaporation and plant transpiration and improve the soil moisture; (3) In the periphery area of the wetland, infiltration should be promoted to ensure the source of water supply to wetlands, which would maintain the existence of wetlands and give full play to their ecological functions. In addition, this study analyzed the relationship between soil moisture and plant diversity of 35 species. 16 plants are considered to be suitable for planting in core area of sandy wetland and 9 plants for planting in the desert hinterland outside the wetland.*

Keywords— **Desert area; Sandy wetland; soil moisture; Zoning protection; Protect the catchment desert.**

I. INTRODUCTION

Wetland is the kidney of the earth, and sand wetland is the main landscape type to maintain the ecosystem of the sand area [1-2]. Wetlands in sandy areas are mainly distributed along both river banks and low-lying areas, which is the ecological front to prevent desert invasion. Studying the source of water and the characteristics of soil water movement is the top priority in studying the sustainable existence of the ecosystem and the function of preventing sand invasion in the sand area [3].

Over the years, researchers have conducted experimental studies on the existence of wetlands in sandy areas, the ecological functions of wetlands and the movement of soil water in wetlands from different angles. Most studies focus on ecological and hydrological processes in sandy areas, and the large scale research is carried out from the hydrological process characteristics of groundwater and surface water. The conclusions are relatively consistent, vegetation is considered in arid areas to have a role in water accumulation [4], and the scale of landscape ecology significantly impacts ecological hydrology in semi-arid regions [5-6]. For instance, Chen [7] indicated that the existence of Tamarix changed the process of precipitation redistribution, which promoted the precipitation infiltration and maintained a high soil moisture. The critical value of ecological water demand in arid areas and the relationship between natural vegetation and groundwater are also studied through experiments [8-10]. The development and utilization of water resources in arid regions of Northwest China are expounded from the levels and zones of river hydrology and ecosystem [11-12]. Heihe River Basin in Northwest China has the most extended history, the most profound depth and the most investment in the study of human development of water resources and its impact on local ecology in arid areas which suffers severe ecological problems [13-15].

The study on the environmental and hydrological coupling of wetlands in the core area of arid desert is different from the hydrological and ecological characteristics of desert edge areas and desertification areas. Taking Ningxia as an example, researchers analyzed the environmental evolution process of the reduction of surface water area and soil moisture in the Aeolian sand area of Ningxia Hui Autonomous Region from the perspective of geological history. 264 towns are ecological divided for hydrological regionalization and the results showed that the environmental water shortage in the middle and north of Ningxia is severe [16-17]. The above studies have been carried out from the overall aspects of ecology and hydrology and less from soil moisture variation in desert areas. Some researchers have done some research on relevant factors, studied the correlation and response-ability between soil moisture and shrub in the desert area [18-22], while discussing the interaction between nitrogen fertilizer and soil moisture in desert area [23-24]. The role of desert shrubs in soil water replenishment and their significance in hydrology and earth system science have also been studied [25].

In summary, studying the sandy wetland is inseparable from water resources. However, the sandy wetland is a unique ecosystem in the desert with relatively isolated eco-hydrologic characteristics, so the hydrologic process and ecological characteristics are different from other wetlands and watersheds. Habahu National Wetland Nature Reserve of Ningxia Hui Autonomous Region is selected as the research area to study the water movement characteristics of the soil profile. Firstly, the catchment area of the wetland is divided according to the catchment area of the wetland. Starting from the overall arrangement and taking the poster height and topographic characteristics as the layout basis, the special method of arranging monitoring points by elevations is implemented, which adopts the method of assimilating extensive data and establishing a large number of monitoring points. On this basis, utilizing microcosmic monitoring, stratified sampling is carried out on the typical soil profile of special parts to test the difference in soil moisture and then study the characteristics of soil water movement at different altitudes and topographic positions in the wetland area. This study would provide a scientific basis for the ecological protection of wetland and the differential protection of other regions.

II. MATERIALS AND METHODS

In August 2021, the soil sampling sites were arranged in Haba Lake Nature Reserve, Yanchi County, Ningxia Hui Autonomous Region (Fig. 1). The selected area is about 144 km^2. The selected sampling sites are based on different altitude gradients, the lowest site is 1424 m, and the highest site is 1591 m above sea level. The landscape sections existing in various ecosystems were taken as the research object. Combined with traditional statistics, geostatistics and GIS, the selected study area was put in a unified latitude and longitude grid, and the 418 selected sampling sites were regularly laid out based on GPS. The altitude interval is 10 m, and the interval of sampling sites at the same altitude is 1 km. The soil samples are collected and tested for moisture. A total of 155 effective soil samling sites were obtained (Table. 1, Fig. 2). Meanwhile, we identified plant species at each effective soil samling site.

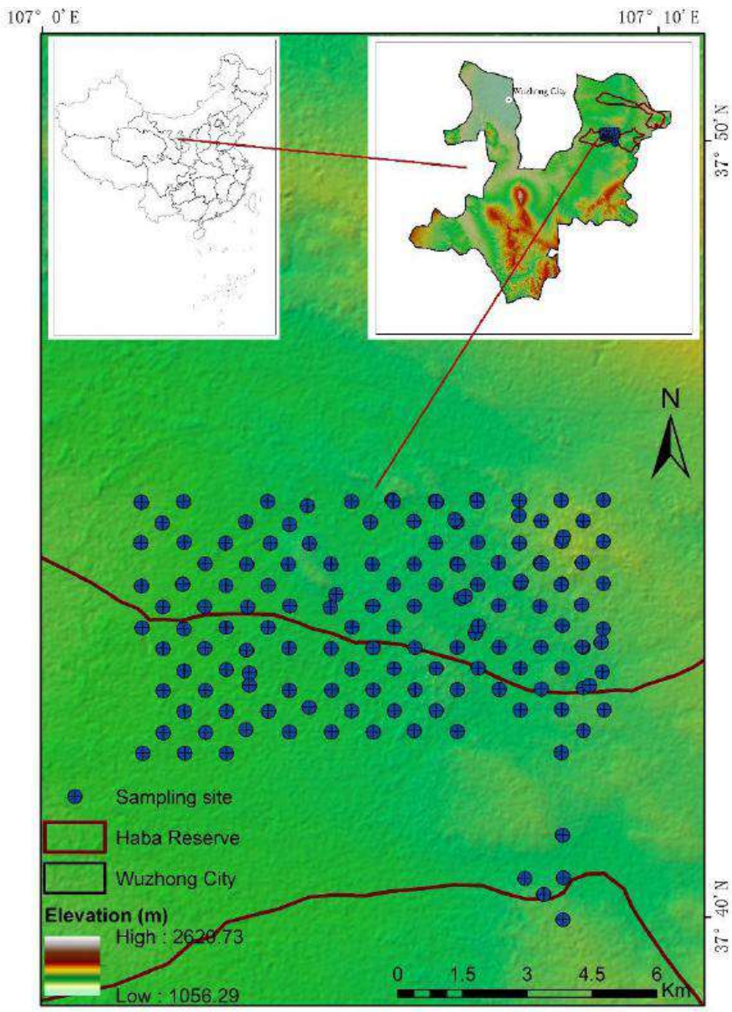

Fig.1 Location map of sampling sites

According to the local quicksand coverage thickness, geomorphic morphology and the characteristics of the underlying terrain, the depth of the soil profile sampled by this research is 120 cm from the surface. Soil samples are collected every 20 cm from 0–120 cm depth for 6 samples in each soil profile. Seal the sample after sampling to avoid moisture evaporation during transportation. After drying in an oven with a design temperature of 105 ° C for more than 1 day, the soil moisture is calculated by weighing. Among the 155 monitored points, 930 soil samples should be collected theoretically. Due to the limitation of soil layer thickness at some sampling points, a total of 837 soil samples were finally collected.

Table 1 Distribution of samples arranged by altitude gradient

No.	Elevation (m)	Number of soil profiles	Soil profile monitoring site (No.)	Selected monitoring site
1	1424—1434	4	393,183,198,408	393
2	1435—1445	8	153、378、168、169、363、199、108、184	184
3	1446—1456	14	333、348、139、154、109、349、364、124、394、125、140、185、334、397	333
4	1457—1467	11	170、350、110、335、141、155、126、380、336、351、	170

			395	
5	1468—1478	10	365、410、127、171、156、366、200、201、352、381	366
6	1479—1489	8	186、337、411、367、128、203、143、202	143
7	1490—1500	12	353、413、382、158、338、396、131、129、189、368、397、147	189
8	1501—1511	29	144、205、371、205-2、173、132、146、339、340、356、398、160、161、354、206、206-2、204-2、190、383、399、414、415、415-2、72、130、73、159、204、385	72
9	1512—1522	28	175、191、370、401、385-2、162、372、384、174、177、357、400-2、133、145、187、369、386、358、387、358-2、176、355、400、177-2、88、343、103、118	133
10	1523—1533	11	148、149、162、373、416、416-2、282、358-2、353-3、373-2、207	282
11	1534—1544	8	388、192、134、402、164、178、178-2、402-2	402
12	1545—1555	0	0	
13	1556—1566	7	417、209、209-2、58、179、208、208-2	58
14	1567—1577	1	403	403
15	1578—1588	2	193-2、193	193
16	1589—1591	2	194、418	194

Note: a steep cliff between 1545m and 1555m altitude, so there are no monitoring sites.

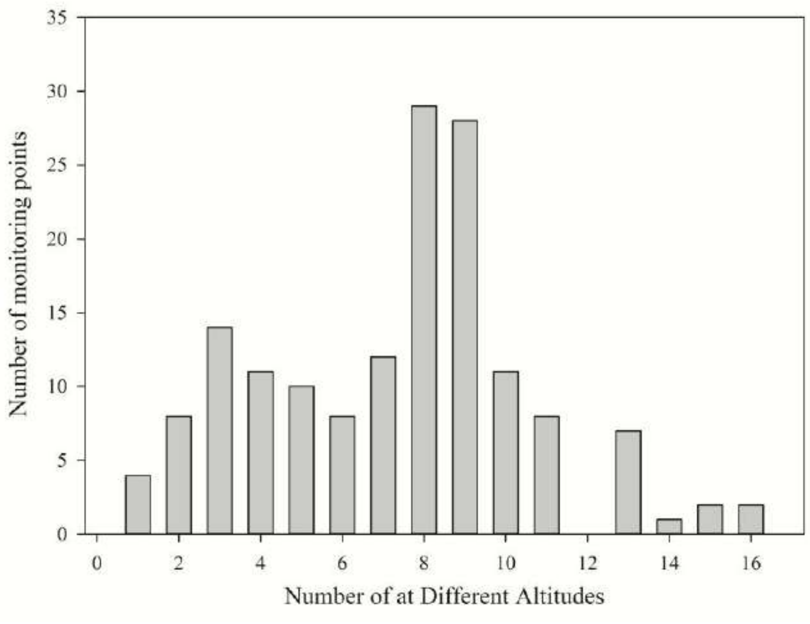

Fig.2 Distribution of soil monitoring points

III. RESULTS

A site with the full collection of soil profile samples was selected in each altitude range to draw a curve of soil moisture distribution along the profile, with a total of 15 curves. The soil profile moisture distribution characteristics at the 15 sampling sites can be divided into rapid evaporation and slow infiltration types (Fig. 3, Fig. 4). And Table 2, Table 3 show the mathematical fitting models.

3.1 *The changing trend of soil moisture below 1500m altitude*

When the altitude is below 1500 m, soil profile moisture evaporates slowly in the surface layer and then rapid infiltration (Fig. 3). The surface soil moisture is the largest, the maximum value is 23.72%, and the minimum value is at the 100 – 120 cm layer, which is only 5.85%. 7 fitted models of soil profile moisture under 1500 m are in Table 2. Among the seven soil profiles, the changing trends of soil moisture at different layers of five soil profiles are similar, with an obvious inflexion point between 20 cm and 40 cm. Only two curves show abnormal changes. The soil profile moisture evaporates rapidly on the surface while the soil moisture increases when the soil depth is below 40 cm at the lowest elevation site. This sampling site is in a typical wetland area, with high surface soil moisture and good biological growth. A dry soil layer appears between 20 – 40 cm depth, which is the typical development characteristic of soil profile of wetland in the sandy area. Soil water sources comes from two parts in sandy area, surface catchment and groundwater supply, with the boundary depth between 20 cm and 40 cm.

From 1490 to 1500 m altitude, the change of soil profile moisture belongs to the characteristics of rapid water infiltration where covering large sand thickness. This site is in the transition area of typical wetlands to sand-covered slopes. The precipitation recharge is rapid infiltration and cannot absorb groundwater recharge due to the slope gradient, so its curve is more similar to soil profile moisture distribution characteristics of sandy soil. At the transition zone of 1490-1500 m altitudes, the three-step coefficients of the fitting model are negative, and it is a seeping type curve. At the elevation of 1424-1434 m, because it is located at the lowest altitude site in the wetland, the soil moisture of the surface soil is very high. The terrain position determines it, so the three-step coefficient of the fitting model is negative as well.

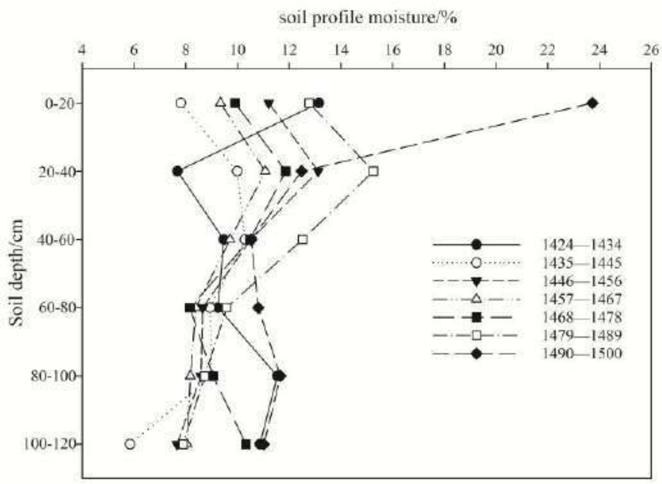

Fig. 3 Variation curve of soil profile moisture with depth from 0 – 20 to 100 – 120 cm at the 1424 – 1500 m altitude

Table 2 Fitting models of soil profile moisture with depth at different altitudes (1424 – 1500 m)

Altitude（m）	Fitting models	R^2
1424—1434	$h = -0.347w^3 + 4.115w^2 - 14.27w + 23.42$	0.852
1435—1445	$h = 0.024w^3 - 0.745w^2 + 3.808w + 4.854$	0.902
1446—1456	$h = 0.198w^3 - 2.151w^2 + 5.830w + 7.618$	0.891
1457—1467	$h = 0.176w^3 - 1.940w^2 + 5.724w + 5.524$	0.908
1468—1478	$h = 0.287w^3 - 2.911w^2 + 8.136w + 4.552$	0.842
1479—1489	$h = 0.306w^3 - 3.376w^2 + 9.493w + 6.598$	0.954
1490—1500	$h = -0.544w^3 + 6.859w^2 - 27.18w + 44.38$	0.992

Note: h represents soil depth, and w represents soil moisture.

3.2 The changing trend of soil moisture above 1500m altitude

The eight soil profile moisture changing curves in Figure 4 belong to the rapid evaporation type. The water infiltration are stable. Except for an abnormal point of surface soil moisture is 24.77 %, the minimum value is only 4.14%, and most values are below 10 %, while the soil moisture in the 30 % of the layers is below 6%. The soil moisture changing process of the 8 soil profiles shows a tremendous decline rate. The soil moisture decline rate of the topsoil layer to the bottom layer can reach more than 10%. The eight curves show obvious boundary inflexions at 60 - 80 cm depth. The layer above the inflexion point is dominated by rapid evaporation, while below the inflexion point is dominated by stable infiltration. According to the moisture content coefficients of the various curves in Table 3 curve, it is also fully in line with the characteristics of rapid evaporation and stable leakage. the laws of the fitting coefficients of the six curves are basically the same except for 1534 – 1544 m and 1567 – 1577 m curves.

Compare Figure 3 and the curve fitting model of Table 2 with Figure 4 and Table 3, the variation trend of soil moisture with depth is obviously different. The curves in Fig. 3 show the sandy soil moisture characteristics of the desert wetland with slow evaporation at the surface and then rapid infiltration below the depth of 40 cm. The curve in Figure 4 shows the soil moisture characteristics that rapid evaporation and rapid infiltration occur at the same time.

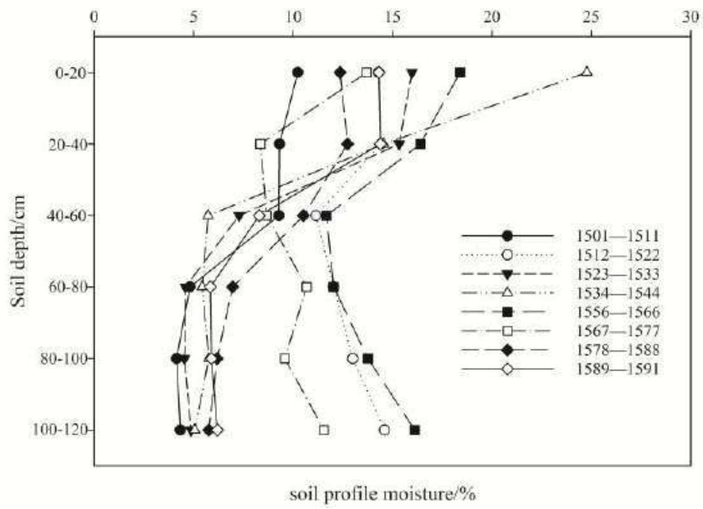

Fig. 4 Variation curve of soil profile moisture with depth from 0 – 20 to 100 – 120 cm at the 1501 – 1591 m altitude

Table. 2 Fitting models of soil profile moisture with depth at different altitudes (1501 – 1591 m)

Altitude (m)	Fitting models	R^2
1501—1511	$h = 0.228w^3 - 2.345w^2 + 5.453w + 6.711$	0.936
1512—1522	$h = 0.073w^3 - 0.341w^2 - 0.747w + 15.62$	0.725
1523—1533	$h = 0.286w^3 - 2.347w^2 + 1.869w + 16.77$	0.938
1534—1544	$h = -0.332w^3 + 4.993w^2 - 24.63w + 45.14$	0.995
1556—1566	$h = 0.053w^3 + 0.287w^2 - 4.794w + 23.21$	0.904
1567—1577	$h = -0.253w^3 + 3.215w^2 - 12.05w + 22.47$	0.789
1578—1588	$h = 0.252w^3 - 2.617w^2 + 6.161w + 8.66$	0.983
1589—1591	$h = 0.265w^3 - 2.330w^2 + 3.247w + 13.60$	0.935

3.3 Different soil depth soil water content distribution characteristics

According to the root extension depth and the actual data of each monitoring and sampling site, the depth of the soil 0 - 60 cm is analyzed in three layers (Fig.5-a, Fig. 5-b, Fig.5-c). The soil moisture in the research area is mainly concentrated on the surface layer of 0-20 cm, and the average value is 14.12 %. The linear regression curve in Figure 5-a show growing trends. The soil moisture at a depth of 20 - 40 cm mainly revolves around 11.92 %. The linear regression curve shows a decreased trend as the altitude increases in the 20 – 40 cm layer. At the soil depth of 40 - 60 cm, that is, the bottom part of the plant root system, the soil mosisture of all monitoring samples has decreased significantly, mainly distributed at about 9.75 %. The depth of 60 cm is a critical division of the surface water and groundwater supply. From a-c in Figure 5, the soil moisture on the three sides of the soil is decreasing with the increase in soil depth, and the decreasing rate is larger at low elevations.

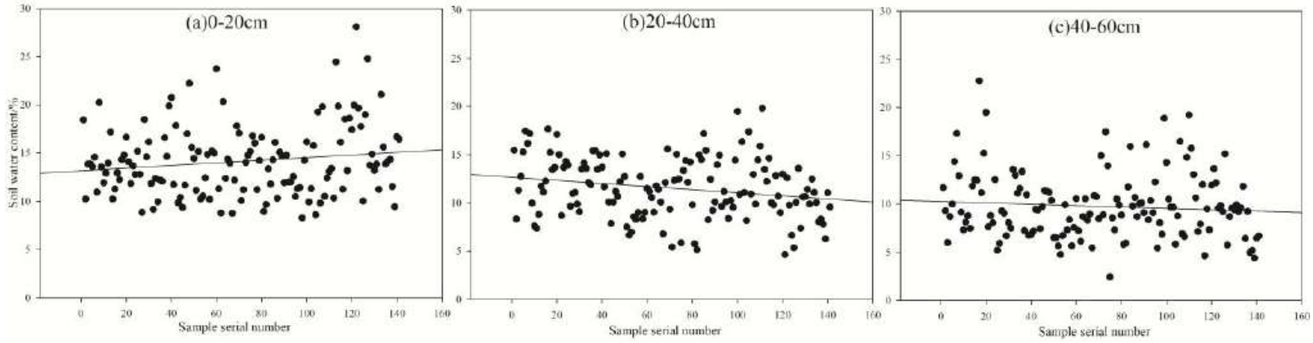

Fig. 5 The distribution map of soil moisture in the soil depth of 0-20cm(a), 20-40cm(b), 40-60cm(c)

3.4 Relationship between surface soil moisture and plant diversity

A total of 35 plant species were identified at the sampling sites, of which 9 species were evenly distributed and grew at the soil moisture of 7 – 28% (Fig. 6-a). 17 plant species are facing degradation problems, these plant species were only collected at less than 6 sampling sites (Fig. 6-b). Pennisetum centrasiaticum and Cynanchum thesioides are suitable for planting in in the desert hinterland, which are mostly distributed in the area the water soil moisture was lower than 14.26% (Fig. 6-c). In addition, 7 plant species were suitable for growing in areas with high soil moisture, but the growth of Heteropappus altaicus and Leymus secalinus were limited when the soil moisture was higher than 19% (Fig.6-d).

IV. DISCUSSION

The annual average precipitation of Haba Lake National Wetland Nature Reserve in Ningxia is 296 mm, which decreases from southeast to northwest with a significant variability rate. More than 80% of the precipitation is concentrated in May-September. Annual evaporation is 2131.7 mm, 6-7 times of precipitation. Figure 7 is the variation of monthly evaporation in Haba Lake National Nature Reserve from 1981 to 2010. In such a severely arid area, the existence of surface wetland and the soil moisture characteristics mainly depends on two aspects: the first is the geological structure, which affects the characteristics of groundwater recharge; The second is the catchment characteristics of the ground and the ability of surface plants to reduce evaporation [26-27]. Therefore, the distribution of soil moisture in soil profiles can be divided into two types: rapid evaporation and rapid infiltration. Desert catchment recharge over a large area is one of the primary wetland sources because of little rainfall and large evaporation capacity on the surface of the wetland. In the transition zone of slope land around the wetland, the thickness of sand cover on the slope surface gradually decreases from the top to bottom, and precipitation rapidly seeps down. The change of soil moisture in Figure 4 at 1534 - 1544 m altitude is a typical rapid seepage process curve in desert area.

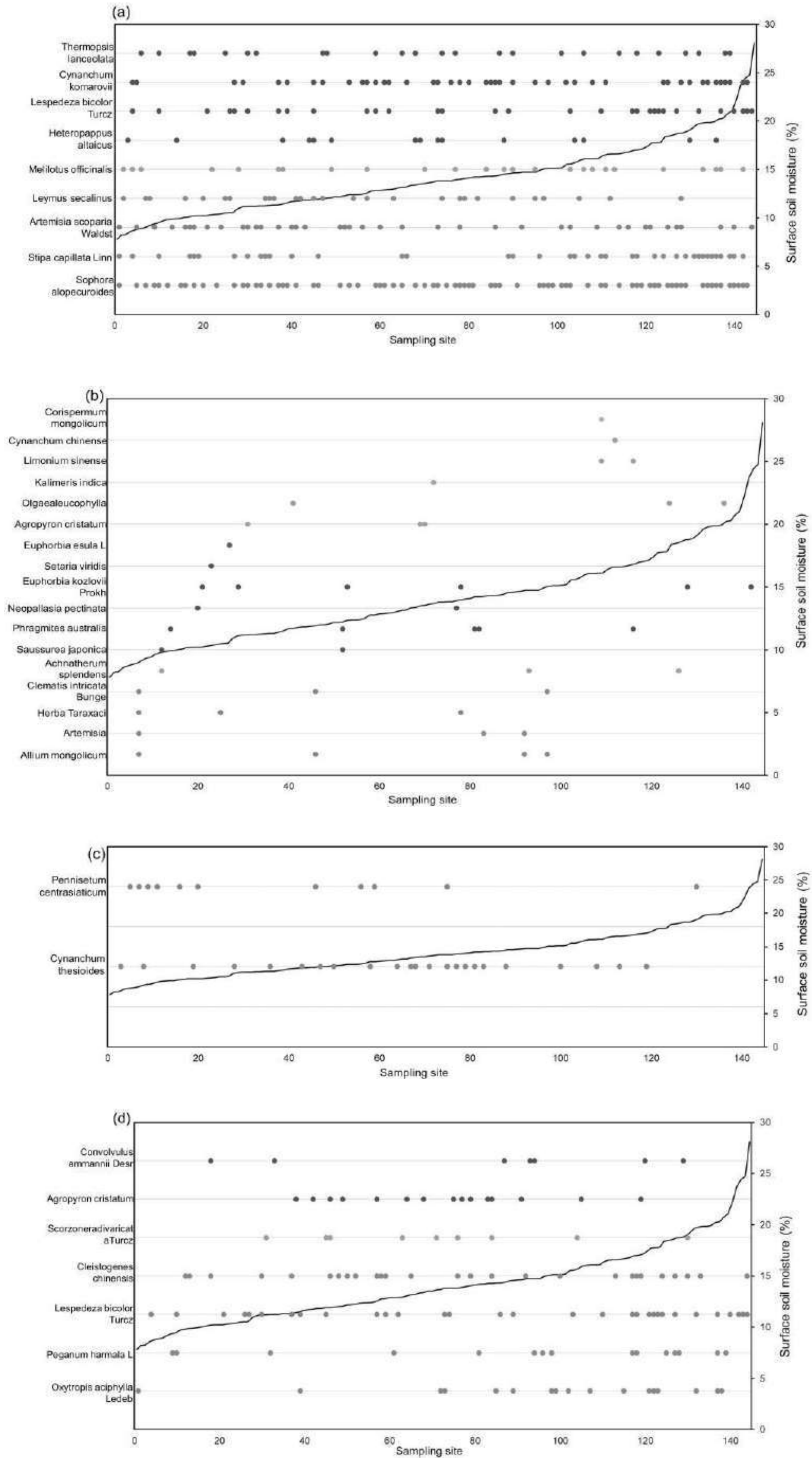

Fig. 6 Plant diversity at different surface soil moisture

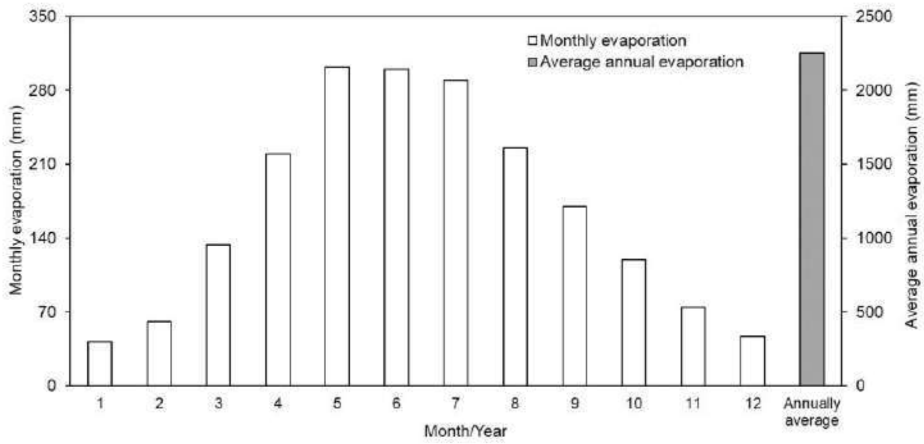

Fig. 7 Variation of monthly evaporation in Haba Lake National Nature Reserve

Wetland hydrology has always been considered an essential driving force for shaping the structure and function of wetland systems [28]. As an important factor affecting the wetland environment, the hydrological process controls wetland functions by affecting physical, chemical and biological processes within wetlands, making wetland systems different from terrestrial systems with good drainage conditions and deepwater aquatic system. Wetlands located in the Delta and other coastal areas have a better ability to withstand storms and typhoons. Wetlands located beside rivers play a more significant role in maintaining the water quality of rivers and controlling floods downstream. Desert wetlands can better regulate groundwater and maintain regional water balance [29-31]. The hydrological landscape is classified according to different hydrogeographic conditions. Soil water transport in wetlands at arid and semi-arid areas mainly focuses on hydro-ecology, including ecological environment effects and Simulation of the hydro-ecological process of wetlands [32-33]. Soil water transport of wetlands in desert areas is obviously different from the wetlands in other ecological environment areas [34].

This study revealed the characteristics of water movements in the soil profile in a small ecological environment of wetland in the sandy area with different surface material composition, terrain and vegetation composition, and other . Basically, the study area can be divided into three regions [35-36]: the first is the typical core wetland area, with slow infiltration and evaporation as the main water transport modes in soil profile; The second type is sloping land around the wetland, with different sand cover thicknesses. After precipitation, the water quickly flows down the impervious layer to the bottom of the slope to make up the wetland soil moisture. The third type is a large area of desert around the wetland. The water soon seeps underground after precipitation, along with the structural characteristics of the underlying stratum, into different groundwater systems or relatively low-lying adjacent wetlands.

V. CONCLUSIONS

The water supply of wetland in the sandy area comes from precipitation and groundwater. However, there is little surface precipitation in the sand area. In order to maintain the existence of wetlands, there must be sufficient catchment areas, which are the deserts in the hinterland of the periphery of wetlands. The precipitation in the arid area is far less than the evaporation capacity. The rapid infiltration of precipitation in the desert area is not only the specific ecological function of preserving water and reducing evaporation in the sand area, but also the main role of wetland in the sand area.

Based on the analysis of the monitoring results of this study, we concluded that the water drop-down surface is mainly gathered in the core area of the wetland in the sandy area. In terms of ecological maintenance, it should focus on surface biological protection to reduce evaporation and improve the water holding capacity of the soil. In the sloping areas, engineering and biological measures should be taken to reduce surface evaporation and plant transpiration, and maintain the normal infiltration of soil and confluence to the soil bottom. In the hinterland of the periphery of wetlands, instead of blindly afforestation, the sand fixation and infiltration promotion should be focused on ensuring the source of water supply to the wetland and thus maintain the existence of the wetland and giving full play to its ecological function. This conclusion is a unique view put forward by this study.

Through the analysis of the relationship between soil moisture and plant diversity, 16 plant species are considered to be suitable for planting in the for planting in core area of sandy wetland and 9 plants for planting in the desert hinterland outside the wetland. 7 kinds of plant species are suitable for planting in whole Habahu National Wetland Nature Reserve.

AUTHOR CONTRIBUTIONS

Liping Zhang wrote the main part of the manuscript. Tianyu Sun, Kai Fei and Longzhou participated in the coordination of the study and reviewed the manuscript, wrote parts of the manuscript. Naiping Song planned and carried out part of the experiments, analyzed the results. Xing Wang, Xinguo Yang, Lei Wang and Lin Chen participated in the coordination of the study and reviewed the manuscript. All authors have read and agreed to the published version of the manuscript.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results.

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Traditional Medicine in the health systems of communities: A first exploring in Veracruz State, Mexico

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Abstract— *Veracruz State has been described as one of the richest states in Mexico, in terms of its medicinal herbal diversity at a local and regional level, and its culture of utilization of such resources in traditional medicine. These resources are valued by their cultural and economic importance and by their usage in the healthcare system. In this study, we explore the value of Mexican traditional medicine knowledge registered in literature on this subject-books, thesis, and publications in ethnobotany scientific journals- from different localities in Veracruz State. We have found reports of a wide variety of plants used to heal different sufferings such as: cough, pain, healing, inflammation, gastrointestinal problems, cancer and diabetes of people who live in communities of Huasteca and Totonaca regions, Nautla, Capital or Central, Mountains, Los Tuxtlas and Olmeca in Veracruz, Mexico. The wide variety of plants found shows us a general scene of Veracruz State's great cultural richness. Some species commonly reported are Bursera simaruba, Tithonia diversifolia, Oenthera rosea, Bidens pilosa, Heterotheca inulodes. The formulations are tea, infusion, cataplasm or washings of the whole plant, root, blossom, bark, stem, leaves or fruits; they are essential to heal each attended illness by people in their communities. This review show that the traditional use of medicinal plants are of great importance due to their healing properties and contributes to an understanding of their biological activity. Furthermore, it will necessary to conduct further researches to evaluate these plants' effectiveness and safety, and to identify the compounds involved in them.*

Keywords— ***Ethnobotany, Mexican traditional medicine, Medicinal Plants, Therapeutic uses.***

I. INTRODUCTION

For many years, traditional medicine has been a valuable resource in the Mexican health system; particularly, in areas of great diversity of plant species and ethnic groups. According to old records, Mexico's herbal medicinal repertoire is one of the most diverse in the world. It contains numerous native plants, as well as species introduced from Europe, Asia and South Africa after the Spanish conquest in 1519 (García-Guerrero and Blanco-Dávila, 2004). The Mexican traditional medicine knowledge and the therapeutic uses of plants by the people of different regions in Mexico (in tea, infusions, decoctions, cataplasms, rubs, baths, macerations and poultices, among others) is an ancient practice still used throughout the country to treat common infections and disorders (Aguilar et al., 1994). Currently, approximately 3,000 to 5,000 plants (both native and introduced species) are used for medicinal purposes by 62 different ethnic groups throughout Mexico (Aguilar, 1999; Lozoya, 1994; Aguilar et al., 1994).

In Veracruz State, the knowledge about the healing properties of plants has been passed on over many generations (de la Cruz, 1991, Chena, 2013). Several researchers, biologists, botanists, students, geographers and anthropologists have published different studies on traditional Mexican medicine contributing to the creation of a rich ethnobotanical bibliographic collection (Frie et al., 1998; Benítez y Wels, 2010). Such studies describe the most frequently used species and their therapeutic use to treat gastrointestinal, respiratory, urinary and skin infections as well as stomachaches, cancer and diabetes (Martinez, 1984, 1992; Del Angel, 1995; Chena, 2013; Lozoya, 1987; Argueta, 1994), which are some of the main health problems in Mexico (Andrade-Cetto, 1995). This information has been gathered from interviews with healers (*curanderos*), midwives, specialists on medicinal plants and key informants in communities who know native plant species and their most frequent uses to treat different ailments.

This can only give us an idea of the importance of plants in the health systems, although there are no specific data to assess the extent of the global use of medicinal plants; the World Health Organization (WHO) estimates that more than 80% of the world's population routinely uses traditional medicine to meet the needs of primary health care and that much of the traditional treatments involve the use of plant extracts or their active ingredients. Since the late 70's, the WHO defined a *medicinal plant* as any plant species that contains substances which can be used for therapeutic purposes or which active principles can serve as precursors for the synthesis of new drugs (WHO, 1978) and therefore, within the framework of the "Health for all by the year 2000" program, it has promoted the study, documentation and kinetic evaluation of plants used in traditional medicine bridging the gap between traditional and modern medicine (Akerele, 1993; Tiwari 2008). Recently, researchers continue carrying out studies to discover and analyze the bioactive compounds and biological activity of medicinal plants, not only to preserve and record their traditional uses throughout the world but also to evaluate their biological and toxic activities in order to validate their use (Akerele, 1993). These validation and research studies could contribute to the development of new drugs and alternative methods to treat health problems as it has been registered in others studies (Heinrich, 2003; Akerele, 1993). For example, some drugs used nowadays were obtained and synthetized from medicinal plants used by the indigenous peoples of different countries.

In this article we present a specific review of different medicinal plants and their therapeutic uses, according to recent studies carried out in communities in Veracruz State, as well as some examples of phytochemical studies about their possible specific biological activity. Also, we explore the value of Mexican traditional medicine knowledge based on the practices of key informants, herbalists ("*hierberas*") and healers ("*curanderos*") from different localities in Veracruz State, as it has been recorded in the literature on this subject -books, thesis, and publications in ethnobotany scientific journals-. Pharmacological reports available on Mexican medicinal plants were also briefly reviewed. This information provides the basis for future in-depth studies on pharmacology of medicinal plants of Veracruz State, Mexico.

1.1. Geographic background

Veracruz state is located in the South East of Mexico, along the Gulf of Mexico (see Figure 1). There are coastal plains along the coast of the Gulf of Mexico, and beyond the coastal plains, hills and canyons are found. Veracruz State covers an area of 72,815 square kilometers (28,114 square miles). Further inland there is Sierra Madre Oriental, which has different names according to the region it occupies: Sierra de Huayacocotla, Zomelahuacan, Chiconquiaco, Huatusco, and Zongolica. Otontepec or Tantima mountain range is located in the North. The mountain range of "Los Tuxtlas", which is not linked to Sierra Madre Oriental, is located in the South. The highest mountains in the State are El Pico de Orizaba (5,610 meters/18,500 feet above sea level) and Cofre de Perote (4,200 meters/13,860 feet). The ecosystem includes seven volcanoes and several lakes, lagoons, and marshlands. The average temperature is 24°C to 28°C (76°f to 82°f), and sometimes the temperature varies depending on the region.

The State is divided into 212 municipalities and it is the third most populated State in the country with a population of 7.1 million people. Veracruz has 10 regions: Huasteca alta, Huasteca baja, Totonaca, Nautla, Capital o Centro, Las Montañas, Sotavento, Papaloapan, Los Tuxtlas y Olmeca (OECD, 2010). The 212 municipalities are distributed along these regions (Figure 1, INEGI 2010). It also features the third largest indigenous population in Mexico. Veracruz has a huge diversity of flora and fauna, and many of these species are unique to Mexico and cannot be found anywhere else in the world. Veracruz is a place of enormous cultural diversity harboring indigenous traditional communities, such as: Popoluca, Totonaca, Nahua and Huasteca peoples (Toledo, 2001).

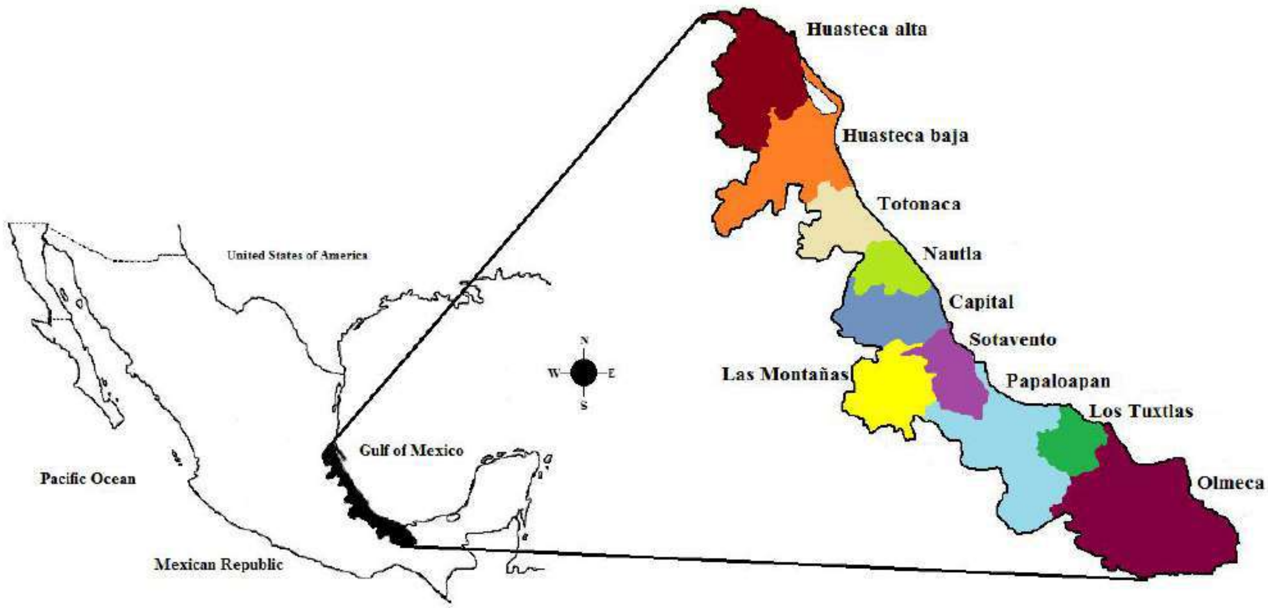

Fig. 1: Geographical location and ten regions of Veracruz State, México (INEGI, 2010).

II. THE VALUE OF MEDICINAL PLANTS IN VERACRUZ STATE

"Mexico is an appropriate place to examine the plant-human richness relationships because of the great degree of diversification of cultures and biota (the study of flora and fauna) in comparison to the rest of the Americas and the world" (Bye, 1993). There are over 5000 species of plants used medicinally by several indigenous groups in Mexico. There are specific records describing the customs and experiences in the preparation of home remedies using medicinal plants of some communities in the region of Veracruz (Cano, 1997a, 1997b). Some of these records were primarily compiled in the Zongolica mountains (Navarro and Avendaño, 2002), in a Galera Nahuatl community of Tantoyuca, Ver., (Del Angel, 1995), in communities of Coscomatepec and Orizaba (Gálvez y De Ita 1992); in four localities of Ixtaczoquitlán municipality (Hernández, 2006); in Tlalnehuayocan (Cabrera-Aguilar, 2010), in Tlalchy, in the municipality of Ixhuacán de los Reyes (Chena, 2013; Enriquez-Lopez et al., 2014), in "Los Tuxtlas" region (Leonti, 2002) and community of Nahua de la Sierra de Santa Marta, Los Tuxtlas (Calatayud, G.A., 1990). Also, there are registers of ethno-botanic studies where the value and importance of using medicinal plants are described. For example; "Algunas Plantas Conocidas como Tóxicas en la Planicie Costera del Norte de Veracruz" published in 1974 by UNAM. Del Amo's study in 1979, where the book "Plantas Medicinales del Estado de Veracruz", was published, registering about 544 medicinal plants used in different locations of Veracruz state.

Considering the distinct regions of Veracruz State, in the North zone called Huasteca Alta (21°22" North latitude, 97°44" West longitude); Bautista (1995) reported 82 plants of medicinal usage in the community of Chinampa de Gorostiza, Veracruz. Also, Del Ángel (1995) elaborated a work in a Nahuatl community of Galera's congregation, which belongs to Tantoyuca's municipality, Veracruz. Relevant information was compiled in here about the knowledge of herbal traditional medicine, getting ethnologic and botanical information of 112 species, related to 52 Families and 100 genuses. The most representative Families are Compositae, Lamiacea and Rutaceae. In this study, Del Ángel (1995) reported plants analgesic, anti-diarrheic, anti-inflammatory, healing, and dermatological properties, among others.

In Huasteca baja region, Smith-Oka (2007) reported a preliminary study of plants used by totonacos doctors, in this place they heal a variety of uneasiness, including; usual illnesses like flu, stomachache, gynecological pains, fractured or strained limbs, related syndromes to culture (including "fright" necaxantle, out of place organs, "malos aires" (when you feel full of air or dizzy, but you have not breathed so much or you are not sick. etc.) and ritual cleanings. Most of the plants were collected in specialist's healing domestic gardens, around 150 specimens were collected, which include 33 Families among them are:

Asteraceae, Euphorbiaceae and Solanaceae. However, some of them such as: *Harpalyce arborescens*, *Adiantum tenerum*, *Cedrela mexicana*, *Croton soliman*, *Bursera simaruba* and *Protium copal*, were obtained from corn plants, from the grass areas destined to cattle or from the little spots of forest that still persist.

In Totonaca region, a plants catalogue used by traditional doctors of Pukuchin Healing House, a huge important herbarium can be seen (about 115 species) to different sufferings (Anemia, wounds, burns, blows, gastric ulcer, inflammation, infection, cough, parasites, diarrhea, muscular pains, diabetes, skin fungus, etc.) as well as their preparation: bathroom, tea, maceration, ointment, cataplasm, just to mention (De los Santos and Ruiz, 2009). Also, there is a study carried out by Olarte (1998) where a bibliographic investigation was done about ethnobotany and phytochemistry of medicinal plants of Filomeno Mata's municipality. (Papantla's mountain range). This research gathered information about 48 plants of medicinal use, documenting usual names, ethnobotanical description, anthropological data and medicinal uses.

To Nautla's region, Montoya (1996) reported 372 useful species, which belong to 286 genuses, in the municipality of Misantla, Veracruz. Each one of the registered species had information about its usual name, usage, section used and the way of preparation. 14 categories were registered, being the medicinal usage the most important aspect, followed by the edible and ornamental usage.

Veracruz state central region, Villamil and Avendaño (1992) presented an internal report called "Aspectos Generales del Uso de las Plantas en Xico, Veracruz". Also Bravo (1993) carried out an ethnobotanical study to the sellers of Naolinco, Veracruz., reporting 299 species, from these ones, 82 were informed to be as medicinal plants of great importance. The most representative Family was Compositae and the most representative uneasiness was the stomachache. On the other hand, Contreras (1996) reported around 350 species distributed in 83 Families were considered medicinal plants used in the central zone of the state of Veracruz. Beaurregard (1996) found in his work "Las plantas útiles expedidas en el mercado de Coatepec, Ver." 112 species of medicinal plants, belonging to 57 Families and 100 genuses; the most representative Families were Asteraceae, Lamiaceae and Lauraceae; and he found the most sold plants are used to treat digestive problems. Santiago and Romero (1996) reported to the municipality of Jilotepec, Veracruz, 399 plant species, which 135 were plants of medicinal utilization. Compositae Family was the most representative and the intestinal infection was the most representative illness. As well, Grapain and Arcos (1999) carried out a study about medicinal plants in the municipality of Jilotepec, Veracruz; registering around 130 genuses and y 63 Families of plants, from which 83 have multiple uses, 16 parts were used in 12 ways of use.

In the mountains region, Martínez-Bolaños (2012) reported about 70 medicinal plant species belonging to 20 Families; the most representative were Compositae, Solanaceae, Labiatae and Verbenaceae, in Barrio de Santa Cruz, Municipality of Tequila in the mountain range of Zongolica. He found 82 categories of multiple usages, among them are the medicinal plants used to heal stomachaches, cough, swelling, diarrhea, healing, skin ulcers and fungus, infections, etc.

While, Grapain López in 2020, reported medicinal plants of the Herbario XALU "Arturo Gómez-Pompa", realized by several authors. She Grapain collected data from the flora medicinal plant of the state of Veracruz, whose species are in the collection of the herbarium XALU "Arturo Gómez-Pompa", integrating 100% of the copies in a format of greater affordability and accessibility, so that people know the collection as well as the uses medicinal of it. Of the 2,205 records, 137 families, 544 genera and 919 species were counted. The 919 species were counted, 12% of the species correspond to the Compositae family, 8% to the leguminosae family, 5% to the Solanaceae family, 4% Lamiaceae family, 4% Euphorbiaceae family, 4% Malvaceae family, 3% Family Rubiaceae.

Henrich et al., (1998) reported culture importance about of medicinal plants in the Sierra de Zongolica of Veracruz State (is part of the Sierra Madre Oriental); where the medicinal plants most frequently mentioned serve toward the treatment of gastrointestinal and dermatological conditions. These two categories yielded the largest number of individual use-reports. For example, the principal species used to treat dermatological problems by Nahua are *Phyllamthus niruri*, *Lobelia laxiflora*, *Heterotheca inuloiedes*, *Bryophyllum calcinum*, *Mercadonia procumbens*, *Stellaria nemorum* and *Tithonia diversifolia*. Particularly, Mexican "Arnica" or Mexican "sunflower" -*Tithonia diversifolia* is commonly used by different groups and also is known from various regions of Veracruz for treating dermatological conditions (Del Amo, 1979). This plant is native to the lowlands of southeastern Mexico and Central America but it is not well known in the ethnobotanical literature in Mexico (Heinrich, 1996).

In Tuxtla's region, Veracruz., Garrido (1997) carried out a study focused on vegetable species used as medicinal by the settlers of eight rural communities of Catemaco, Veracruz. In this research, it was reported about 104 species used by settlers have different medicinal usages and the most predominant illnesses were gastrointestinal.

The most representative Families were Compositae, Euphorbiaceae and Labiatae. Mendoza-Márquez (2000) reported in his ethnofarmacological-chemical study of the perennial high jungle of Los Tuxtlas, Veracruz, (Tropical Biology Station of los Tuxtlas Reserve) the knowledge and traditional use of medicinal plants. In here were found 309 species of 91 Families and 218 genuses of medicinal use plants. The best representative Families are Asteraceae (34 species), Piperaceae (16 species), Euphorbiaceae (14 species), Rubiaceae (14 species) and Fabaceae (13 species). Some species of major representative in medicinal traditional use are *Sambucus mexicana*, *Bursera simaruba*, *Asclepias curassavica*, *Momordica charantia* and *Piper auritum* (for further information, look up details in these studies, quoted references and published articles).

In this ethno-botanical studies conducted in these communities it is possible to observe the different uses and the value or importance of medicinal plants in some regions in Veracruz State. Many of them are important medicinal resources which continue to be used nowadays and others are hardly ever used (Heinrich, 1989). Oftentimes some of these medicinal plants or parts of them (roots, leaves, etc.) are sold in popular markets, important easy-to-find places for their sale and distribution. These popular markets known as *mercados*, are widely known in Mexico and other Latin American countries (González-Stuart, 2010). In markets we can also find stalls known as *hierberías* where people sell a variety of herbal products. Here, herbs are usually sold by weight, dried or cut in small pieces or in small packets (Bellucci, 2002).

In general, popular markets have a larger selection of crude herbs as these products cannot be found in most of the more modern stores or supermarkets. Thus, popular markets in Mexico retain some of the cultural flavor regarding medicinal herbs traditionally used for healing and usually have herbal vendors who may also act as healers (Farnsworth et al., 1985; Taddei-Bringas et al., 1999; Gheno-Heredia et al., 2011; Bellucci, 2002).

Studies allowed investigators to conclude the existence of a basic group of 1,000 plants which have been used in Mexican traditional medicine almost throughout the whole country for nearly 400 years. Plant based treatments continue to play an essential role in primary health care (Aguilar et al., 1994; Balick and Cox, 2003), and it has been estimated that in Mexico, about 25% of the population still depends on the use of medicinal plants.

2.2. Medicinal plants and their properties in traditional healing practices

Historically all medicinal preparations derived from plants are different or similar among communities. In Veracruz state there are different sorts of preparations and the therapeutic use of plants is a common practice; the different preparations of plants reported, using part of the plant (e.g. its fruit) or a combination of its parts (leaves, bark, roots, fruit and flowers), or combining plants of different species which suggests the presence of chemical agents that are effective against health problems. Thus, from a scientific point of view, it is essential to establish the relationship between the empirical use of plants and their real biological and therapeutic activities.

We analyzed the ethno-botanical literature available and we found common species as well as the traditional medicinal knowledge of some regions of Veracruz State. In Table 1 and 2 we can see a large amount of plants and their uses registered by some communities of Veracruz State. In this table it is important to observe that the people used the plants for treatment as well as anti-inflammatory, wounds and burns, pain, arthritis, rheumatism, antiseptic, respiratory problems, gastrointestinal and genital problems, cancer and diabetes. The formulation in tea, infusion and washings is more common in all communities evaluated in this paper.

For instance, we observed that the species *Thithonia diversifolia* is used to treat skin rashes and *Tagetes erecta* L., to treat wounds and pain, both are of the Family Asteraceae and were found in the community of Tlalchy, Ixhuacán de los Reyes, Ver., Totonacas, Ver., and San Pedro Soteapan, Veracruz, respectively. The species *Rhoeo discolor* L is used to treat wounds and washes, it belongs to Family Commelinaceae it was found in the community of Tlalnelhuayocan, Ver., and San Pedro Soteapan, Veracruz. Oenothera rosea L´Her. Ex Aiton was found in the communities Tlalnelhuayocan, Ver., Tlalchy, Ixhuacán de los Reyes, Ver., Barrio de Santa Cruz and Tequila, Ver., this species are used for wounds, bruises and burns, anti-inflammatory.

The *Sedum praealtum* and *Sedum purpurens* of the Family Crassulaceae are used for mouth sores and sores, respectively; and the species *Nicandra physaloides* and *Solanum nigrum* are used to treat wounds; they belong to Family Solanaceae they were found in the community of Tlalchy, Ixhuacán de los Reyes and Nahuatl Galera, Tantoyuca, Veracruz. Also, were found important different species of Family Gesneriaceae, Labiatae, Onagraceae and Rutaceae in this communities (see Table 1). This information on the therapeutic uses of medicinal plants is based on the species registered by each community and region (see Table 1 and 2, for some examples); which it shows that there are several plants with different uses and sometimes they are used for the same condition. Therefore, in the future, the species reported in Table 1 will require scientific studies to verify their biological effectiveness.

Also, it will be known their chemical composition and the effective dose for each ailment and treatment. Only a relatively small number of Mexican plants have been studied for their possible medical applications and even a smaller number of them have been studied for their efficacy, safety, active ingredients and the type of extracts contained in them.

Table 1. Therapeutic uses, part (s) used and formulations of plants in three different localities of the State of Veracruz: Huasteca, central or capital and Los Tuxtlas, Ver.

Family[b]	Scientific name [b]	Local/Common name [a, b]	Therapeutic uses [a, c]	Part (s) used [a, b]	Formulation [a, b]	Region / Locality	Ref.
						Region: Huasteca Alta	
Aloeaceae	Aloe vera L.	Sábila	Burns	Leaves	Direct, macerated,	Galera Nahuatl Community, Tantoyuca, Ver.	Del Ángel, 1995.
Crassulaceae	Sedum purpurens L.	Balsámica	Sores	Leaves	Macerated and rubbed on skin		
Rubiaceae	Hamelia patens Jacq.	Maduro Zapote	Wounds	Leaves	Boiled for washes		
Solanaceae	Solanum nigrum	Hierba mora	Wounds	Leaves	Compresses with boiled leaves		
Sterculiaceae	Guazuma ulmifolia	Guacima	Wounds	Cambium	Applied on affected area		
						Region: Capital or Central	
Anacardiaceae	Amphipteryngium adstringens Schiede ex Schlecht.	Cuachalalate	Skin infections and swelling	Bark	Tea	Tlalnelhuayocan, Ver.	Cabrera-Aguilar, 2010
Chenopodiaceae	Chenopodium graveolens Willd.	Epazote zorrillo	Postpartum baths, cramps, abdominal cramping, dewormer, allergies, chickenpox, measles, diarrhea	Flowers, leaves and stems	Tea		
Commelinaceae	Rhoeo discolor (L'Hér.) Hance ex Walp	Maguey morado	Bruises and wounds. Used to stop bleeding	Leaves	Washes	Tlalnelhuayocan, Ver. San Pedro Soteapan, Ver. Zoque-Popoluca.	Cabrera-Aguilar, 2010. Muñoz, 2012.

Compositae	Árnica montana L.	"Árnica"	Vaginal douches, infections, swelling, wounds	Flowers, leaves and stems	Infusions and washes	Tlalnelhuayocan, Ver.	Cabrera-Aguilar, 2010.
Compositae	Artemisia cina Berg ex Poljakov	Hierba maestra	Bile, hemorrhoids, itchy neck, stomachaches (adults), diarrhea	Leaves	Tea and washes		
Compositae	Eupatorium pycnocephalum Less	Hierba pico de pájaro"	Cancer, diarrhea	Flowers, leaves and stems	Tea		
Compositae	Eupatorium sp.	"Zacanene"	Labor induction	Leaves	Tea and washes		
Compositae	Heterotheca inuloides Cass	Árnica Arribeña	Wounds, swelling, infections	Flowers, leaves and stems	Tea and washes		
Compositae	Taraxacum officinale F.H. Wigg.	Diente de león	Wounds	Flowers, leaves and stems	Washes		
Compositae	Verbesina turbacensis H. B. & K	Árnica de árbol	Wounds	Leaves	Washes		
Equisetaceae	Equisetum hyemale L	Cola de caballo	Fever, constipation, kidney diseases, urinary tract infections	Leaves and stems	Tea		
Euphorbiaceae	*Acalypha alopecuroidea* Jacq	Chinaguatillo, hierba del cáncer, comalillo"	Bruises, sores, kidney diseases, wounds, pimples, sore throat, stomachaches	Flowers, leaves and stems	Washes and tea		
Gesneriaceae	*Kohleria deppeana* (Schltdl. & Cham.) Fritsch	Tlalchichinole, chinol	Mouth sores, cough, vaginal washes,	Leaves, stems and roots	Baths, washes, tea, chewed		

			gastritis				
Gramineae	*Cymbopogon citratus* (DC.) Stapf	Zacate limón	Bile, nerve diseases, influenza	Leaves	Tea, infusion	Tlalnelhuayocan, Ver. Tlalchy, Ixhuacán de los Reyes, Ver.	Cabrera-Aguilar, 2010. Chena, 2013.
Labiatae	*Marrubium vulgare* L.	Hierba del vaporrub	Cough, asthma, stuffed nose, respiratory system (infections)	Leaves	Tea	Tlalnelhuayocan, Ver.	Cabrera-Aguilar, 2010.
Labiatae	*Rosmarinus officinalis* L.	Romero	Anti-inflammatory, postpartum baths, relapses, magical-religious significance, wounds, disinfectant, labor induction, stomach lotion, fever	Leaves	Tea, baths, washes		
Labiatae	*Salvia microphylla* H. B. & K	Mirto	Magical-religious significance, postpartum baths, dewormer, hemorrhoids, worms in babies, headache, wounds, skin injuries, earache	Flowers, leaves and stems	Tea, baths, washes Ointment	Tlalnelhuayocan, Ver. Tlalchy, Ixhuacán de los Reyes, Ver.	Cabrera-Aguilar, 2010. Chena, 2013.
Malvaceae	*Malva parviflora* L.	Malva	Vaginal discharges, postpartum baths, fever, vaginal douches	The whole plant	Tea, baths, washes	Tlalnelhuayocan, Ver.	Cabrera-Aguilar, 2010.
Melastomataceae	*Arthrostema* *Ruiz* & Pav	Mocachane de siembra	Relapses, postpartum baths, fever,	Flowers, leaves and	Tea, washes		

			wounds, influenza	stems			
Myrtaceae	*Psidium guajava* L.	*guayaba*	Stomachaches and diarrhea	Leaves and fruits	Tea	Tlalnelhuayocan, Ver.	Hernández-Suárez, 2016.
Onagraceae	Oenothera rosea L'Hér. ex Aiton	Hierba del golpe	Wounds, bruises and burns (skin injuries), strains, anti-inflammatory, gastritis.	Leaves, stems and fruits	Tea, washes, infusion or plasters.	Tlalnelhuayocan, Ver. Tlalchy, Ixhuacán de los Reyes, Ver.	Cabrera-Aguilar, 2010. Chena, 2013.
Plantaginaceae	Plantago australis Lam.	Pata o manita de león, llantén	Wounds, venomous animal bites, eye pain	Flowers, leaves and stems	Baths, washes	Tlalnelhuayocan, Ver.	Cabrera-Aguilar, 2010.
Rosaceae	Rubus fagifolius Schltdl. & Cham	Mora de monte	Vaginal infection, blood purifier	Flowers, leaves and stems	Washes, tea		
Rutaceae	Ruta graveolenes L	Ruda	Stomachache, magical-religious significance, hemorrhoids, bile, postpartum baths, gastritis, abortive, swelling, blood circulation.	Leaves and flowers	Tea and baths		
Selagenillaceae	Sellaginella aff. galeotti	Doradilla, flor de piedra, garrapatilla	Epileptic seizures, kidney diseases.	Leaves	Tea		
Urticaceae	Urtica chamaedryoides Pursh	Ortiga, Chichicastle	Kidney diseases, anemia, arthritis, rheumatism	Leaves and flowers	Tea		
						Region: Capital or Central	
Asteraceae	Brickellia	Prodigiosa	Bile	Leaves and	Infusion	Tlalchy, Ixhuacán de los	Chena,

	cavanillesii			stems		Reyes, Ver.	2013.
Asteraceae	Cebollina	Ageratum af. houstonianum	Wounds, scrapes, sores and burns, used as a healing agent	Flowers, leaves and stems	Direct application of infusion or washes		
Asteraceae	Tithonia diversifolia	Gigantón	Skin rashes	Leaves	Infusion		
Asteraceae	Aldama dentata	Mozote amarillo	Gastrointestinal problems	Flowers, leaves and stems	Infusion		
Asteraceae	Bidens pilosa	Mozote blanco	Gastrointestinal problems such as swelling	Flowers, leaves and stems	Infusion	Tlalchy, Ixhuacán de los Reyes, Ver. Totonaca, Ver.	Chena, 2013. De los Santos and Ruiz, 2009.
Asteraceae	Eupatorium af. pichichense	Hierba negra	Skin injuries such as cuts, scrapes and bruises	Leaves and stems	Infusions and washes	Tlalchy, Ixhuacán de los Reyes, Ver.	Chena, 2013.
Caprifoliaceae	Sambucus nigra ssp. canadensis	Sauco	Cough	Flowers, leaves and stems	Infusion		
Clusiaceae	Hypericum sp.	Hierba del tapón	Diarrhea	Leaves and stems	Infusion		
Crassulaceae	Sedum praealtum	Siempreviva	Mouth sores	Leaves	Infusion		
Davalliaceae	Polypodium rivulare	Petatillo	Used to prevent influenza relapses	Flowers, leaves and stems	Infusion		
Gesneriaceae	Moussonia sp.	Bayetilla	Gastrointestinal problems	Flowers, leaves and stems	Infusion		
Gesneriaceae	Moussonia sp.	Bayetilla	Gastrointestinal problems	Flowers, leaves and stems	Infusion		
Labiatae	Ocimum selloi	Hierba del zopilote	High blood sugar	Flowers, leaves and stems	Infusion		

Lauraceae	Litsea glaucescens	Laurel	Skin injuries	Leaves	Infusion or ointment		
Lythraceae	Cuphea nitidula	Hierba del ángel	Used to prevent influenza relapses	Flowers, leaves and stems	Infusion		
Magnoliaceae	Magnolia dealbata	Magnolia	Wounds	Leaves	Ointment		
Onagraceae	Lopezia racemosa	Hierba del sarampión	Measles	Flowers, leaves and stems	Direct application of infusion		
Papaveraceae	Bocconia frutescens	Gordolobo	Cough	Leaves and stems	Infusion		
Rutaceae	Casimiroa edulis	Zapote blanco	High pressure.	Leaves	Infusion		
Solanaceae	Nicandra physaloides	Belladora	Wounds	Flowers, leaves and stems	Ointment		
Umbelliferae	Foeniculum vuilgare	Hinojo	Influenza	Leaves and stems	Infusion		
Rutaceae	Citrus limón	Limón	Influenza, sores and wounds.	Fruits	Infusion, diluted for washes.	Tlalchy, Ixhuacan de los Reyes, Ver. Comunidad Nahuatl Galera, Tantoyuca, Ver.	Chena, 2013. Del Ángel, 1995.
						Region: South Veracruz, Los Tuxtlas.	
Amaryllidaceae	Bauhinia divaricata	Lirio	Skin fungus	Flower and stems	Washes	San Pedro Soteapan, Ver. Zoque-Popoluca.	Muñoz, 2012.
Aquifoliaceae	Ilex belizensis	Palito verde (Tsus kuy)	Wound healing	Leaves and stems	Washes		
Asclepiadaceae	Artemisa ludoviaciana Nutt	Estafiate (Poma´ay)	Dewormer, cough, stomachaches, infections	Leaves	Tea and infusion.		
Asteraceae	Tagetes erecta L.	Cempazuchil (Tsus mooya)	Wound healing, erysipelas, diarrhea,	Leaves	Washes and Braches		

			earaches. Magical-religious significance				
Bombaceae	Pachira aquatica	Apompo (Uakta)	Diabetes and cystitis	Leaves	Tea and infusion		
Bromeliaceae	Aechmea bracteata	Kardón (kardun)	Earaches and cystitis	Leaves	Tea and infusion		
Burseraceae	Bursera simaruba	Palo mulato (Ts_k)	Diarrhea, dysentery, fever, wounds and gastritis. Used as a living fence	Leaves and stems, braches	Tea and washes	San Pedro Soteapan, Ver. Zoque-Popoluca. Totonaca, Ver.	Muñoz, 2012. De los Santos and Ruiz, 2009.
Cactaceae	Epiphyllum crenatum	Pitahaya (Ñuchtyi)	Used to prevent miscarriages and to stop bleeding. Also used for food	Leaves and stems	Tea and infusion		
Cochlospermaceae	Cochlospermum vitifolium	Pongolote (Puts kuy)	Snake and black widow bites, anti-inflammatory and diabetes	Leaves	Tea	San Pedro Soteapan, Ver. Zoque-Popoluca.	Muñoz, 2012.
Curcubitaceae	Mormodica charantia L.	Bejuco condeamor (cundeamor)	Heals skin wounds, rashes, diabetes, scabies	Leaves and Braches	Wash wounds, decoction of leaves as a refreshing bath. Root decoction as tea		

Information gathered from key informants. [a]

Information gathered from a literature review. [b]

Information gathered from key informants but interpreted by the authors of this article. [c]

Table 2. Example of some species of medicinal plants used by regions of Veracruz State.

Region	**Scientific name (Family)**	**Local/Common name**	**Therapeutic uses**	**References**
Huasteca Totonaca Mountains	Aloe vera	Sábila	Stomachache, cough, blows, healing.	Del Ángel, 1995. Martínez-Bolaños, 2012.

Huasteca Totonaca	Hamelia patens Jacq. (Rubiaceae) Hamelia erecta	hierba negra Bayetilla	Bath, wounds.	Del Ángel, 1995. De los Santos and Ruiz, 2009.
Totonaca Olmeca	Bursera simaruba (Burseraceae)	Palo mulato, Chaca (TAZUN)	Diarrhea, dysentery, fever, wounds and gastritis.	Muñoz, 2012. De los Santos and Ruiz, 2009.
Totonaca Mountains	Heterotheca sp (Compositae) Heterotheca inulodes (Compositae)	Árnica	Blows, healing, genital infections, gastritis, ulcer gastric.	De los Santos and Ruiz, 2009. Martínez-Bolaños 2012.
Capital or central, Mountains	Sambucus nigra var canadienses (Caprifoliaceae)	Sauco	Cough, fright.	Chena, 2013. Martínez-Bolaños, 2012.
	Oenothera rosea (Onagraceae)	Hierba del golpe	Wounds, bruises and burns (skin injuries), strains, anti-inflammatory, gastritis, break down, kidney pain, blows, fractures, bruises.	Cabrera-Aguilar, 2010. Chena, 2013. Martínez-Bolaños, 2012.
Capital or Central, Totonaca	Titothonia diversifolia	Gigantón	Skin rashes, dandruff, scabies, and baldness.	Cabrera-Aguilar, 2010. De los Santos and Ruiz, 2009.
	Taraxacum officinale FH Wigg. (Compositae)	Diente de león	Wounds, kidney stones, intoxication, toothache, acne.	
	Bidens pilosa (Asteraceae)	Mozote blanco	Anemia, gastrointestinal problems such as swelling.	De los Santos and Ruiz, 2009. Tlalchy
Central or Capital, Olmeca	Rhoeo discolor (L´Her.) Hance ex Walp (Commelinaceae)	Maguey morado	Bruises and wounds. Used to stop bleeding.	Cabrera-Aguilar, 2010. Muñoz, 2012.
Mountains, Totonaca	Plantago mayor (Plantaginaceae)	Lanté	Kidney pain, organ inflammation and internal ulcers.	Martínez-Bolaños, 2012. De los Santos and Ruiz, 2009.
Mountains, Central or Capital	**Solanum nigrescens (Solanaceae)**	**Hierba mora**	**Break down, wounds.**	**Del Ángel, 1995. Chena, 2013.**

2.3 Phytochemical studies and biological activity of medicinal plants

Phytochemical studies are necessary to understand the pharmacological and biological activities of the different species of plants used in traditional medicine, to gain an understanding of their mechanisms of action, and for quality control purposes. Currently, more than ever before, there is information available on the active principles of some plants. The reason is that modern detection techniques and research have greatly expanded our knowledge about some of the possible medicinal and toxicological properties of plants (Awang, 2009). Some of these studies have shown that plants have a limitless ability to synthesize aromatic substances, mainly secondary metabolites, of which at least 12,000 have been isolated, a number estimated to be less than 10% of the total (Mallikharjuna, et al., 2007). Some secondary metabolites may be involved in plant odour (terpenoids), pigmentation (tannins and quinines), and flavor (Capsacin) (Mallikharjuna, et al., 2007). These molecules give plants their medicinal value which is appreciated by people, because of their vital role in human healthcare and for their economic value (Fazal et al., 2012). For example, diseases caused by microorganisms, the increasing resistance in many common pathogens to currently used therapeutic agents, such as antibiotics and antiviral agents, has led to a renewed interest in the discovery of novel anti-infective compounds (Fazal et al., 2012). Thus, the natural products derived from medicinal plants have proven to be an abundant source of biologically active compounds, many of which have been the basis for the development of new pharmaceuticals.

Therefore, the valuable medicinal properties of different plants are due to the presence of several constituents i.e. saponins, tannins, alkaloids, alkenyl phenols, glycoalkaloids, flavonoids, sesquiterpenes lactones, terpenoids and phorbol esters (Tiwari and Singh, 2004). These active compounds of interest present in the medicinal plants have continuously been of great concern to the scientists (Ahmad et al., 2012). Among them, some act as synergistic agents and enhance the bioactivity of other compounds. Thus, the presence of these phytochemicals in several medicinal plants could be responsible for the observed effects in traditional medicine.

Therefore, plants have shown to be a potential source new antimicrobial, antifungal, anti-inflammatory or analgesic agents (Clark and Hafford, 1993). Some species cited in the table 1 have been evaluated by other authors, John-Dewole and Oni, 2013 and Hinojosa-Dávalos et al., 2012, reported the presence of saponins in the leaves of species *Thitonia diversifolia* and *Tithonia tubaeformis*, and confirmed that they have anti-inflammatory, antifungal and antiparasitic properties; Owoyele et al., (2004) shown Antiinflammatory and analgesic properties of *Tithonia diversifolia* leaf extract.

The antioxidant and hepato-protective effects of *Heterotheca inuloids* have been reported too and this species have mutagenic/anti-mutagenic dual effect mediated by quercetin (Ruiz-Pérez et al., 2014). On the other hand, *Rhoeo discolor* ethanolic crude extract evaluated in an *in vitro* system, showed anti-mutagenic, anti-genotoxic and anti-oxidative activities (González-Ávila et al., 2003) and anti-tumoral activity of aqueous crude extract of Rhoeo discolor (Rosales-Reyes et al., 2008). We could also confirm the reason why some medicinal plants are used to treat several treatments or affections. The presence of various active ingredients revealed by phytochemical screening and biological activity supports the resourcefulness of some plant extracts reported by researchers in other countries, and some of these medicinal plants are found in Veracruz state (Table 1 and 2). Therefore, all these natural agents (medicinal plants) reported in this paper, could to be a potential source for new compounds or drugs with diverse novel mechanisms of action and to treat infectious diseases, as have been recorded by others authors.

III. CONCLUSIONS

In this review, we show that Veracruz State has a wide variety of medicinal plants, which functions as a health care system for several communities. Also, it was observed that plants continue to be an important resource to treat different health problems in Veracruz State and all over Mexico. Furthermore, all the information collected in this review also shows that there is an important variety of plant species with different uses and applications. The formulation in tea and washes is more common in all communities evaluated in this paper. In addition, several of the species registered in different parts of Veracruz State are used for the same healing purposes or the same species can be used for various medicinal purposes. The phytochemical and ethno-pharmacological studies reviewed confirm the found properties in some plant species reported by the state of Veracruz. Therefore, the existing literature and experiments could be a successful step in the development of new drugs and be an alternative to cure certain diseases in Veracruz, Mexico. Also, from a scientific point of view, it is important to continue carrying out studies to confirm the efficacy and safety of medicinal plants, as well as to discover potential healing properties and to identify the compounds involved in the medicinal plants used by different communities of the ten regions in Veracruz, Mexico.

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Quality control of pine needles extracts (PNE) by using electroanalytical methods

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***Abstract*— *pH represents the concentration of free H^+ in pine needles extracts (PNE) and is therefore an important initial parameter in quality control. Electrical conductivity and pH of samples of fresh and stored for 20 days of PNE with black cumin oil and olive oil had values of 0.00 due to the encapsulation of water molecules, pH and electrical conduction was not possible. The pH of the other samples was in a weakly acidic environment because the pH of natural pine needles is 3.8. Electrical conductivity values in all samples except pine needle extract and honey increased during storage. By monitoring the parameters of pH and electrical conductivity in the quality control of PNE, it gives us a significant insight into the physical state of the phases and the way of storage.*

***Keywords*— electrical conductivity, extracts, PNE, pH, quality control**

I. INTRODUCTION

Koutsaviti et al. 2022 reported that pine needles has many physiological, pharmacologial activities and it is a good source of antioxidants and phenolic compounds including phenolic acids, flavonoids, procyanides [1], [2]. Pine trees (*Pinus densiflora*) have shown a wide range of biologically functional activities and many methods have been applied for phenolic compounds extraction from pine trees. Increasing interest has been devoted to naturally occurring compounds polyphenols, because of their beneficial health effects [3].

In this research, natural extraction agents such as: honey, black cumin oil (*Nigella sativa*), ethanol, olive oil and traditional jam (pekmez) were used to extract ions and bioactive components from pine needles. Analyzes of pH and electrical conductivity of fresh extracts and after 20 days of storage in a dark place were performed. There is growing interest in the antioxidant activities of plant extracts such as that obtained from pine needle. To obtain these antioxidant compounds, an extraction process is necessary. Extraction is a procedure performed to obtain metabolites in plants such as alkaloids, phenolics, flavonoids, glycosides, and others using selective solvents [4].

Honey is one of the most appreciated and valued natural products introduced to humankind since ancient times [5]. It is a by-product of flower nectar and the upper aero-digestive tract of the honey bee, which is concentrated through a dehydration process inside the bee hive [6]. The main nutritional and health relevant components are carbohydrates, mainly fructose and glucose but also about 25 different oligosaccharides [7]. Honey plays an important role as an antioxidant, anti-inflammatory, anti-bacterial agent and augments the adherence of skin grafts and wound healing process [8].

Black seed or black cumin (*Nigella sativa*), which belongs to the Ranunculacea family, is an annual herb with many pharmacological properties [9]. Black cumin is native to a vast region of the eastern Mediterranean, northern Africa, the Indian subcontinent, and Southwest Asia, and is cultivated in many countries, including Egypt, Iran, Greece, Syria, Albania, Turkey, Saudi Arabia, India, and Pakistan [10]. Black seed and its main active constituent, thymoquinone, to be medicinally very effective against various illnesses including different chronic illness: neurological and mental illness, cardiovascular disorders, cancer, diabetes, inflammatory conditions, and infertility as well as various infectious diseases due to bacterial, fungal, parasitic, and viral infections [11]. The aqueous and oil extracts of the seeds have been shown to possess antioxidant, antiinflammatory, anticancer, analgesic and antimicrobial activities.

Ethanol has been widely applied as a viable solvent [12]. Park et al. 2022 reported that using ethanol at a concentration of 1000 μg/mL showed good electron donating ability among the pine needle extracts [13].

The olive tree is a relatively small evergreen tree, with narrow silvery leaves and small white flowers, known for its longevity [14]. Olive oil is characterized by a high proportion of monounsaturated oleic acid, but the main peculiarity of extra-virgin oil is the presence of remarkable quantities of phenolic compounds, notably hydroxytyrosol and oleuropein [15]. High consumption of extra-virgin olive oils, which are particularly rich in these phenolic antioxidants (as well as squalene and oleic acid), should afford considerable protection against cancer (colon, breast, skin), coronary heart disease, and ageing by inhibiting oxidative stress [16].

Jams are delicious and nutritious spreads typically made from fruit, sugar and pectin that ensure availability of fruits in off-season [17]. The apple jam (traditionally called pekmez) can be defined as a concentrated and shelf-life extended form of apple juice produced by boiling without adding sugar or any other food additives [18]. Pekmezs are especially rich in carbohydrates and minerals, also pekmezs contain vitamins, antioxidants and flavonoids [19]. There are two different kinds of pekmez, either liquid pekmez or solid pekmez [20].

II. EXPERIMENTAL PART

Instrumentation

The following instrumentation was used in this research: pH meter GLP 21 CRISON with a resolution of 0.1, 0.01, 0.001, conductometer GLP 31 CRISON.

Methods

Preparation of PNE

Pine needles extracts (PNE) were prepared by adding 20 g of pine needles accurately weighed on an analytical balance ± 0.0001 g to 100 mL of a natural extraction agent (honey, ethanol, black cumin, olive oil, apple jam/nigella sativa). .

Further electroanalysis of the PNE prepared in this way was performed immediately and after 20 days of storage in a dark place.

Fig. 1: Pine needle extracts (PNE)

Determination of pH

The measurement procedure with a combined glass electrode is simple, and involves carefully immersing the electrode in the extract, whereby the instrument measures the potential difference, i.e. the pH value. Before using the combined glass electrode, it is necessary to calibrate the instrument with standard buffer solutions of exactly known pH value.

Exactly 10 g of pine needle extract with each individual natural extractant was weighed on an analytical balance with an accuracy of 0.0001 g. Then the combined glass electrode was immersed and the pH value was measured. Three parallel measurements were performed for each sample.

Determination of electrical conductivity

The platinum electrode measurement procedure is simple, and involves carefully immersing the electrode in the extract, whereby the instrument measures the electrical conductivity expressed in μS/cm. Before using the platinum electrode, it is necessary to calibrate the instrument with standard buffer solutions of exactly known pH value. As with the measurement of the pH value, the conductivity of all samples was measured in three parallel measurements using the same procedure.

III. RESULTS AND DISCUSSION

Table 1. shows the results of determining the pH and electrical conductivity (κ, µS/cm) of various natural extraction agents.

*Table 1***.** Results of pH and electrical conductivity (κ, µS/cm) of various natural extraction agents

Natural extraction agents	pH	κ, µS/cm
Honey	0,00	2,61
Ethanol	6,99	0,17
Black cumin	0,00	0,00
Olive oil	0,00	0,00
Apple jam/Pekmez	4,79	0,00

Based on the results, it can be seen that pure honey, nigella sativa and olive oil have a pH value of 0.00. The highest pH value was observed for pure ethanol and that value was 6.99, while the slightly lower pH value was for jam and was 4.79. Slightly different results were observed for electrical conductivity, (κ, µS/cm), and honey had a conductivity of 2.61 µS/cm, while ethanol had a conductivity of 0.17 µS/cm. The values of pH and electrical conductivity (κ, µS/cm) for the samples that had values of 0.00 are actually a consequence of their viscosity. Namely, with less viscous samples, it is much more difficult to measure pH and conductivity with electrodes. *Table 2.* shows the results of determining the pH and electrical conductivity (κ, µS/cm) of various natural extracts of pine needles immediately after maceration.

Table 2. Results of pH and electrical conductivity (κ, µS/cm) of various freshly prepared PNE

PNE	pH	κ, µS/cm
Honey	4,44	11,52
Ethanol	5,39	14,25
Black cumin	0,00	0,00
Olive oil	0,00	0,00
Apple jam/Pekmez	3,87	0,00

The pH value of the extract of honey and pine needles was 4.4, while the conductivity of this mixture was 11.52 µS/cm. A slightly higher pH value was 5.39 for extract ethanol and pine needles, and the conductivity of this extract was also slightly higher than extract honey and pine needles and that value was 14.25 µS/cm. The values of pH and electrical conductivity were again 0.00 for samples of macarata *Nigella sativa* and pine needles, as well as olive oil and pine needles. According to Palamutoğluthe et al. 2022 oil is the continuous phase in all emulsion formulations and has no electrical conductivity, indicating that the emulsions are water-in-oil emulsions [21]. Which suggests that the results in this research for the mentioned samples that had a pH and electrical conductivity of 0.00 are in accordance with the mentioned phenomenon that the emulsions are water-in-oil emulsions. The pH value of the jam and honey extracts was slightly lower than the pH value of the jam without the addition of pine needles. ***Table 3.*** shows the results of determining pH and electrical conductivity (κ, µS/cm) of various natural extracts of pine needles after 20 days of maceration.

*Table 3***.** Results of pH and electrical conductivity (κ, µS/cm) of various stored PNE

PNE	pH	κ, µS/cm
Honey	3,50	9,48
Ethanol	5,65	39,2
Black cumin	0,00	0,00
Olive oil	0,00	0,68
Apple jam/Pekmez	4,26	60,3

The results of pH and electrical conductivity of the extracts after 20 days of storage showed that the extract of honey and pine needles had a lower pH value and a lower value of electrical conductivity than the freshly prepared extract. However, the values of pH and electrical conductivity of the extract of ethanol and pine needles had higher values compared to the freshly prepared extract. The jam extract of pine needles had a significantly higher electrical conductivity value of 60.3 µS/cm.

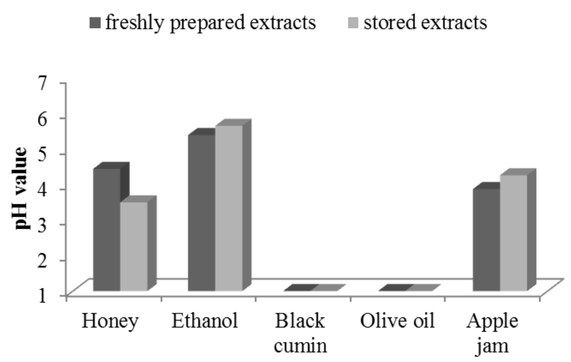

Fig. 2: Results of pH of various freshly prepared and stored PNE

On Figure 1. and Figure 2. the results of the influence of storage time on pH values and electrical conductivity (κ, µS/cm) of pine needle extract are clearly presented.

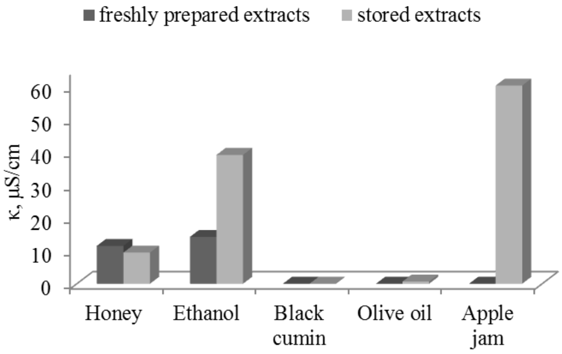

Fig.3: Results of electrical conductivity (κ, µS/cm) of various freshly prepared and stored PNE

IV. CONCLUSION

Pine needle extracts (PNE) can be used as additives in food, pharmaceutical and cosmetic industries. pH and electrical conductivity of black cumin oil was 0,00 in freshly prepared and stored PNE due to the encapsulation of water molecules. In the other samples, an increase in the value of electrical conductivity was observed, which can be concluded that the ions were extracted from the samples during storage.

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Application of papaya leaf extract *(Carica papaya L.)* as a natural insecticide on the larvae of the Aedes aegypti mosquito vector of dengue fever

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Abstract— *The toxicity of the ethanol extract of papaya leaves (Carica papaya L.) on the mortality of the Aedes aegypti mosquito as a vector of dengue hemorrhagic fever. The aim of the study was to determine differences in the concentration of ethanol extract of papaya leaves (Carica papaya L.) on the mortality of Aedes aegypti mosquitoes and the most effective concentration (LC50) for 24 hours of treatment. The study used a completely randomized design with 5 extract treatments, namely: 10 ppm; 50 ppm; 100 ppm; 500 ppm and 1000 ppm and 1 control group with 3 replications. The results of the study were analyzed using one-way analysis of variance. and continued with the BNT test at a significance level of 0.05. To determine the most effective concentration of papaya leaf extract in killing the larvae of Ae. aegypti used Probit Analysis (Finney Method) with Minitab 17 software.The results showed that increasing the concentration of ethanol extract of papaya leaves (Carica papaya L.) would increase the number of test larvae mortality. Based on the results of the probit analysis test, the most effective concentration to kill 50% of Aedes aegipti mosquito larvae was 413.906 ppm (LC50 = 413.906 ppm). Ethanol extract of papaya leaves (Carica papaya L.) has the potential to be developed as a natural insecticide for the Aedes aegypti mosquito vector of dengue hemorrhagic fever.*

Keywords*— *Extract, Papaya leaf, natural insecticide, Aedes aegypti.

I. INTRODUCTION

North Sulawesi in January 2015 was categorized as an Extraordinary Event (KLB), the five-year cycle of the Dengue Hemorrhagic Fever (DHF) outbreak that hit eight districts/cities in North Sulawesi, killing eight people who were positive for the virus transmitted by the Aides aegypti mosquito.

Until now, no vaccine has been found to kill the virus that causes dengue fever. One way to prevent the spread of dengue hemorrhagic fever (DHF) is to prevent transmission of the dengue virus, namely by controlling and eradicating vectors to cut off disease transmission. (WHO, 2005). The World Health Organization (WHO) estimates that every year there are 50-100 million cases of dengue virus infection worldwide.

Fogging is one of the mechanical control methods. Unfortunately, smoking is considered less effective because it tends to repel mosquitoes from their nests, not kill mosquitoes. The chemical method used is the distribution of larvicides such as abate in mosquito breeding places. Indeed, the use of chemical larvicides has succeeded in controlling Aedes aegypti larvae, but the continuous use of chemical larvicides will actually cause resistance and various environmental problems. cause environmental pollution, poisoning, death of non-target organisms, and produce residues.

Due to the negative impact caused by chemical insecticides, it has prompted experts to look for alternatives to vector eradication, namely by using natural insecticides that are safer, easier, cheaper, and do not have a toxic impact on humans. Control using vegetable insecticides or biolarvicides is one of them. Vegetable insecticides made from plants that contain bioactive compounds that are toxic to insects. Organic pesticides are easily biodegradable so they are not harmful to the environment (Octavia, et al. 2008).

The LC_{50} value is a certain concentration that causes the death of the test organism as much as 50%, while the LC_{90} value causes the death of 90% (Klaassen and Watkins 2003)

Plants that have been extracted and isolated by researchers containing active compounds of vegetable insecticides on Aedes aegypti mosquito larvae are soursop seeds (Annona muricata) with LC_{50} = 117. 27 ppm (Komansilan et al. 2012), Hutun seeds (Barringtonia asiatica Kurz) with Lethal Concentration LC_{50} = 35.72 ppm (Komansilan and Suriani. 2016) and lavender leaf (Lavandula angustifolia) with Lethal Concentration LC_{50} = 87.0285 ppm (Komansilan et al. 2021).

Papaya (Carica papaya L.) is one of the plants that can be used as a natural insecticide, because it is effective in controlling insects (mosquitoes). Chemical constituents in papaya leaves include papain enzymes, karpain alkaloids, pseudo-karpains, glycosides, carposids, saponins, saccharose, dextrose, and levulose. (Dalimartha, 2005). Robinson (1998) stated that alkaloids can interfere with the formation of the constituent components of peptidoglycan in bacterial cells, so that the cell wall layer is not fully formed and causes bacterial death.

Flavonoids are polyphenol compounds. Phenol compounds are able to denature protein bonds in cell membranes, so that the cell membrane becomes lysed and the possibility of phenol penetrates into the cell nucleus resulting in changes in cell permeability which can result in inhibition of cell growth or cell death (Peleczar and Chan, 1986).

According to Alboneh (2012), in his research, the substances in papaya leaves which are thought to have potential as insecticides for Aedes aegypti mosquitoes are papain enzymes, saponins, flavonoids, and tannins. The use of natural larvicides can be done to reduce chemical larvicide resistance which is still widely used by the community (Astuti 2009).

II. RESEARCH METHODS

Research Location and Time

This research was conducted at the Integrated Science Laboratory, Faculty of Mathematics and Natural Sciences, Manado State University from May to September 2022.

Tools and Materials

The tools used are: test tube, mosquito cage, blender, filter, rotary evaporator, desiccator, digital scale, measuring cup and micro pipette. Research materials: papaya leaves, ethanol, aquades, Aedes aegypti mosquito larvae, fish feed, paper strain.

Research design

The design used was a completely randomized design (CRD), with 6 treatments and 3 replications, namely: K1 = Control, K2 = .10ppm, K3 = .50ppm, K4 =100ppm, K5 = 500ppm and K6 = 1000ppm

Observation

The parameter observed was the mortality percentage of the Aedes aegypti mosquito, which was calculated using the formula proposed by Kundra (1981):

M = a/b x 100%

Where: M = percentage of mosquito mortality

Ae. aegypti

a = number of mosquitoes Ae. Dead Aegypti

b = number of mosquitoes Ae. aegypti who used.

Work procedures

1. Lava propagation of Aedes aegypti mosquitoes

a) Aedes aegypti mosquito larvae media is made by filling a plastic container with water and the inner wall is lined with filter paper. Filter paper serves as a place for female mosquitoes to attach their eggs.

b) Eggs attached to filter paper are then dried at room temperature and stored in a closed container. For hatching eggs, filter paper is dipped in a plastic tray filled with water and after 24 hours the eggs will hatch and grow into first instar larvae.

c) First instar larvae will develop into second, third (4 days) and IV instar (2 days) instar larvae. Once every 2 days, the larvae were fed 1-2 grams of fish pellets. III/IV instar larvae used in the test.

2. Extract Making

Making papaya leaf extract is as follows:

a) Papaya leaves are separated from the stems, washed, and allowed to dry in the air to avoid sunlight.

b) Dried papaya leaves are mashed using a blender.

c) The mashed leaves were extracted by maceration using technical ethanol until all the components had been extracted.

d) The ethanol extract obtained was evaporated with a vacuum rotary evaporator until thick.

3. Toxicity Test.

a) Provide a solution of papaya leaf ethanol extract in a test tube with concentrations: 10ppm, 50ppm, 100ppm, 500ppm and 1000ppm.

b) In each test tube, 10 larvae of Aedes aegypti mosquitoes are inserted

c) The calculation of mortality was carried out after 24 hours of treatment.

Data analysis

To distinguish the toxicity between treatments of several concentrations of papaya leaf extract against Aedes aegypti mosquito larvae, it was analyzed using one-way ANOVA analysis at a 95% confidence level ($\alpha = 0.05$), followed by the BNT test. Analysis to determine Lethal Concentration (LC_{50}) can be determined through Probit analysis, which aims to determine the concentration of papaya leaf extract (Carica papaya linn) which can kill Aedes aegypti mosquito larvae by 50% of the population tested.

III. RESULTS AND DISCUSSION

1. Comparative Test of Mortality Rate of Ae. Aegepty in Giving Ethanol Extract of Papaya Leaves *(Carica papaya linn).*

The data presents the number of deaths and the mortality rate of Ae. aegepty at five levels of concentration, namely 1000 ppm, 500 ppm, 100 ppm, 50 ppm, 10 ppm, and 0 ppm (control). The data used is mortality rate data (mortality) in the form of percentage scores from 0% to 100%. The 0% figure states that out of 10 mosquito larvae, none have died, while the 100% indicates that all 10 mosquito larvae have died.

The first step before the analysis is carried out is to describe the research variables (descriptive statistics) which includes the presentation of the average value and variation (standard deviation) of each concentration of the ethanol extract of papaya leaves.

Table 1. Description of the Average Value and Variation of Each Concentration of Ethanol Extract of Papaya Leaf Ethanol (Carica papaya linn).

Concentration	The average	Variation
0 ppm (Control)	0.00	0.00
10 ppm	0.00	0.00
50 ppm	1.00	1.00
100 ppm	3.00	0.00
500 ppm	6.67	0.58
1000 ppm	9.33	0.58

Source: Primary Data Processed, 2022

Graphically the data contained in table 1 can be presented as follows:

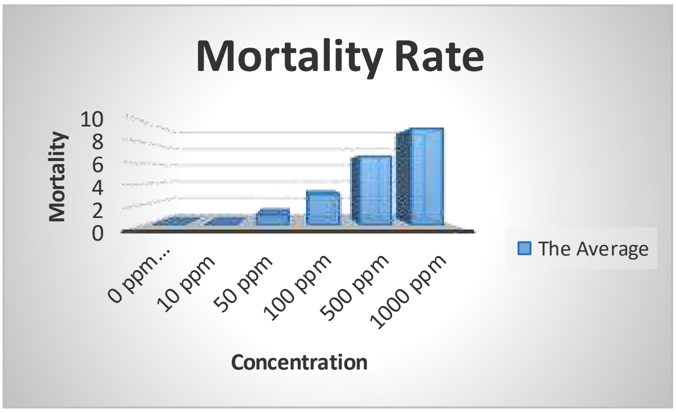

Fig.1. Description of the Average Value and Variation of Each Concentration of Giving Ethanol Extract of Papaya Leaves (Carica papaya linn).

In Figure 1, the height of the bar graph represents the average mortality rate of mosquito larvae for each concentration. From the table and figure above, it appears that there are differences in the mortality rate of Ae. Aegypti at various concentration levels from 0 ppm (control), 10 ppm to 1000 ppm. To find out whether there are differences in the mortality rate of Ae. Aegypti which were significant (significant) at the five concentrations were tested with One Way ANOVA or equivalent to a Completely Randomized Design.

Then, One-way ANOVA was tested. Treatment or concentration of Papaya Leaf Ethanol Extract (Carica papaya linn). declared significant (significantly different) if the value of Fcount > Ftable or the value of Sig F (P-value) < 0.05 (error rate 5%).

Table 2. Results of One-Way ANOVA Concentration Data on Papaya Leaf Ethanol Extract (Carica papaya linn).

Mortality rate					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	224.667	5	44.933	161. 760	,000
Within Groups	3.333	12	,278		
Total	228.000	17			

The test results in table 2 show that the Fcount value is 161,760, and Sig F is 0.000. From the F-Statistics table, it is obtained Ftable of 2.90 Because the value of Fcount > Ftable and Sig F < 0.05 indicates that there are significant differences in the mortality rate of mosquito larvae at various concentrations.

To find out which concentration gives the highest mortality rate, a post hoc test is used, namely the smallest significant difference test (honestly significance difference). given a different notation (different subset), indicating that there is a difference between concentrations.The following are the complete test results:

Table 3. Follow-up Test Using Test (BNT) Concentration Data Giving Ethanol Extract of Papaya Leaves (Carica papaya linn).

Concentration	The average	Notation
0 ppm (Control)	0.00	a
10 ppm	0.00	a
50 ppm	1.00	b
100 ppm	3.00	c
500 ppm	6.67	d
1000 ppm	9.33	e

Note: The same notation shows insignificant differences, while different notations show significant differences.

In table 3 above, it can be seen that given a concentration of 0 ppm (control, or without ethanol extract), the mortality rate of Ae. aegypti at 0.00% or no test larvae will die. With an increase in concentration to 10 ppm, it gives the same test larvae mortality rate (same notation) when compared to a concentration of 0 ppm, which is 0.00%. On the other hand, by increasing the 50 ppm ethanol extract, it gave a higher mortality rate of mosquito larvae (different notations) when compared to concentrations of 0 ppm and 10 ppm, which was 10.%. Meanwhile, by increasing the concentration to 100 ppm in the administration of ethanol extract, it gave the mortality rate of Ae. aegypti which is bigger (different notation) when compared to 50 ppm concentration, with a mortality rate of 30.00%. With an even higher concentration, namely 500 mortality rate of 66.70% and concentration of 1000 ppm, the mortality rate of aedes aegypti mosquito larvae reached 93.30% mortality rate. So it was concluded that the concentration of 10 ppm, 50 ppm, 100 ppm, 500 ppm and 1000 ppm would give the mortality rate of Ae mosquito larvae. aegypti different.

The increase in mosquito mortality was caused by an increase in the concentration of the extract. This indicates that each extract concentration has a different toxic level. It was proved that the low concentration of the extract had a low level of toxicity, causing low larval mortality. On the other hand, with a high concentration of extract, it will have a high level of toxicity, causing high mortality. This is in line with what was expressed by Watuguly (2003), that the factor that most determines the potential danger or safety of a compound is the relationship between the levels of chemical substances and their effects. In addition, the interaction of a toxic substance with biological systems is directly related to the amount of toxic material.

2. Biolarvicide Toxicity Test from papaya leaf extract *(Carica papaya L.)* against Mosquito Larvae Aedes aegypti

The results of the toxicity test of papaya leaf extract against Aedes aegypti mosquito larvae are presented in table 4.

Table 4. The results of the toxicity test of the ethanol extract of papaya leaves (Carica papaya L.) against the larvae of Ae. aegypti after 24 hours of treatment

Extract Type	Concentration (ppm)	Number of dead larvae / 10 head			Percent mortality
		D 1	D2	D3	
control	0	0	0	0	0
Papaya leaf	1000	9	10	9	93.33
	500	7	7	6	66.66
	100	3	3	3	30
	50	1	2	0	10
	10	0	0	0	0

To find out at what concentration papaya leaf extract was most effective in killing the larvae of Ae. aegypti requires a more in-depth analysis tool, namely Probit Analysis (Finney Method) using Minitab 17 software.

The data used as a whole were obtained from 10 larvae of Ae. aegypti in each replication (there were 3 replications) so that 30 larvae of Ae. aegypti as a whole. Table 5 below presents the estimation parameters of the probit analysis model:

Table 5. Parameter estimation model of papaya leaf extract probit analysis on larvae Ae. aegypti

Parameter Estimates		Standard Error	95,0% Normal CI	
Parameter	Estimate		Lower	Upper
Mean	413,906	45,2901	325,139	502,673
StDev	326,504	44,2593	250,325	425,866

Graphically, the probit analysis curve is presented as follows:

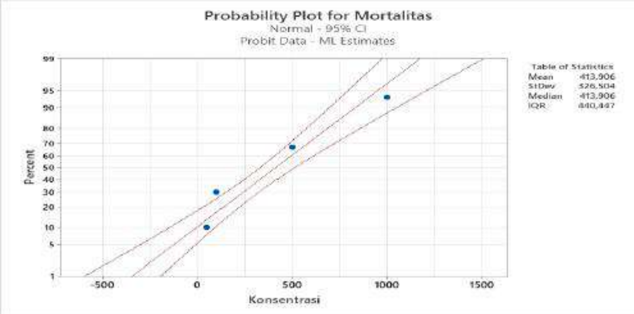

Fig.2. LC_{50} value of papaya leaf extract on Ae. aegypti larvae after 24 hours treatment.

Table 5 and Figure 2 present the LC_{50} or Mean Lethal Concentration values of papaya leaf extract based on the results of Probit Analysis. The test results show the value of the LC_{50} mortality concentration of Ae. aegypti is by giving a concentration of 413.906 ppm. Thus, the concentration figure of 413.906 ppm is the concentration of the most effective papaya leaf ethanol extract to kill Ae. aegypti as much as 50% for 24 hours of treatment. According to the toxicity criteria based on the Australian Petroleum Energy Association (1994) a concentration of 413.906 from papaya leaf extract or (LC_{50} = 413.906 ppm) at 24 hours of observation was included in the criteria for Moderately Toxic

Dyah (2011) explained that the flavonoid compounds contained in papaya extract have various pharmacological activities and have chemical structures that are toxic to pests, this if given in sufficient concentrations it is able to poison pests properly through the digestive process so that plants become healthy in carrying out their activities. absorption of nutrients for vegetative growth processes such as plant height.

Juliantara (2010), reported that papaya leaves contain secondary metabolites such as alkaloids, flavonoids, terpenoids, saponins and other compounds such as papain enzymes which are used as vegetable pesticides.

Pesticide residues cause insect feeding activity to decrease and even stop. In addition, insects also showed a decrease in movement activity. In addition, papaya leaves contain flavonoid substances that work as neurotoxins which are thought to cause aphids to experience a decrease in movement activity. (Dyah, (2011) This is in accordance with Rosyidah (2007), who explained that flavonoid compounds can cause weakness in nerves. as well as damage to the spiracles that result in insects unable to breathe and eventually die.

IV. CONCLUSION

Ethanol extract of papaya leaves (*Carica papaya L.)* has toxicity to the mortality of Aedes aegypti mosquitoes. Based on the test results, the extract concentration of 413.906 ppm was able to kill 50% of Aedes aegypti mosquitoes. The most effective concentration used to kill 50% of Aedes aegypti mosquitoes was 413,906 ppm.

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Assessing the Carbon Footprint and Carbon Mitigation Measures of a Major Full-Service Network Airline: A Case Study of Singapore Airlines

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***Abstract*—*In this qualitative longitudinal case study, Singapore Airlines carbon footprint is examined for the period covering the company's 2010/11 to 2021/22 financial years. The study's qualitative data was examined by document analysis. The case study found that Singapore Airline's carbon footprint is comprised of its Scope 1 direct emissions that are produced from the provision of its passenger and air cargo services together with the carbon dioxide (CO_2) emission produced from the ground service equipment and vehicles used in its ground operations. The carbon footprint also includes its Scope 2 indirect emissions, which are produced from the airline's consumption of electricity at its Singapore hub. Singapore Airlines has implemented a very comprehensive environmental policy and has pledged to achieve net zero carbon emissions by 2050. The largest source of carbon dioxide (CO_2) emissions is from the airline's passenger services. Singapore Airlines has implemented extensive carbon dioxide (CO_2) emissions reduction measures that have focused on the reduction in aircraft weight, and highly efficient fuel management procedures. Other key carbon dioxide (CO_2) emissions reduction measures include improved operational procedures, the optimization of air space management in collaboration with key air traffic control agencies, the use of cleaner energy vehicles, an extensive range of energy efficiency measures in its buildings and facilities, the use of sustainable aviation fuels, and the use of more energy efficient ground power sources. In addition, the acquisition and operation of the state-of-the art, next generation aircraft, such as the Airbus A350-900XWB and the Boeing 787-10 have helped the airline to mitigate its carbon footprint.*

***Keywords*— Airline, carbon dioxide (CO_2) emissions, carbon footprint, case study, Singapore Airlines.**

I. INTRODUCTION

The global air transport industry provides services to virtually every country in the world and has played an integral part in the creation of the world economy. In addition to its economic significance, the air transport industry also provides important social benefits (Daley, 2016; Daley & Thomas, 2016; Rizzi & Rizzi, 2022). As a means of mass transportation, air transport provides the linkage between many different countries and cultures. This contributes to the cultural understanding and increases multicultural cooperation (Caves, 2003; Vespermann & Holztrattner, 2011). Air transport also contributes to sustainable development. By facilitating tourism and global trade, the air transport industry generates economic growth, provides jobs, increases revenues from taxes, and fosters the conservation of protected areas (Air Transport Action Group, 2011, p. 3). Air transport is also very important for long distance travel and enables remote areas to be connected with the rest of the world (Bråthen & Halpern, 2012).

The global commercial airline industry has grown over the past twenty years or so at an annual rate of 4.5–5% in the passenger and 6% in the air cargo segments. Whilst such developments have contributed to the globalization of the economy and overall social welfare, they have at the same time had an increased impact on the environment and society in terms of greenhouse gas (GHGs) emissions. In particular, the further growth of emissions of greenhouse gas (GHG) driven by growth of world air transportation demand could further contribute to global warming and the subsequent climate change (Janić, 2014). This is because carbon dioxide (CO_2) emissions produced from air transportation services have an adverse environmental impact because of their potential greenhouse effects (Postorino & Mantecchini, 2014). Consequently, the global air transport industry's contribution to global carbon dioxide (CO_2) emissions has been under considerable scrutiny since the early 2000s (Bows-Larkin et al., 2016). This increasing focus on the carbon dioxide (CO_2) emissions is because the global aviation industry generates a substantial carbon footprint (Filimonau et al., 2018). Accordingly, the strong historical growth in commercial air transport traffic has increasingly raised concerns over the impact of air transport operations on air quality (Budd, 2017, Daley, 2016; Harrison et al., 2015). This is because the increase in airline flights increases the amount of carbon dioxide (CO_2) that is released into the atmosphere (Fatihah & Abdul Rahim, 2017). In the global airline industry, the carbon dioxide (CO_2) emitted by aircraft engines has become a particular area of concern (Grote et al., 2014), due to their harmful impact on the environment (Olsen et al., 2013). Furthermore, aviation-related emissions are unique in that they occur at higher altitudes as well as from surface-based activities (Olsen et al., 2013). Aviation emissions impact surface air quality on a multiple scale—nearby airport pollution peaks from airport landing and take-off (LTO) emissions, whilst intercontinental pollution is produced from aircraft cruise emissions (Yim et al., 2015). Thus, aviation-related emissions not only impact air quality, but also the climate (Ashok et al., 2017; Kapadia et al., 2016).

The environmental sustainability of air transport has been receiving greater focus in recent times due to its critical impact on climate change and on the environment. Environmental issues associated with the global air transport industry have grown in importance in recent years. in response to these concerns some airlines have been proactive and have developed and demonstrated their "green" credentials (Wu et al., 2018). Indeed, in recent decades, "greening" (ensuring the sustainable development of the global air transport system) has been viewed as a highly significant part of the agenda by almost all the industry's involved stakeholders (Janić, 2011). Many airlines have defined and implemented environmental related strategies that are designed to "green" their operations (Çabuk et al., 2019; Chan et al., 2021; Migdadi, 2020), or where possible make them more environmentally friendly. At a global level, the peak global airline industry body – the International Air Transport Association (IATA) – have recognized the requirement to address the global challenge of climate change and the organization has subsequently adopted a set of ambitious targets to mitigate carbon dioxide (CO_2) emissions from air transport operations. The association has targeted an average improvement in aircraft fuel efficiency of 1.5% per year from 2009 to 2020 (International Air Transport Association, 2022b). At the associations annual general meeting in 2021, the airline industry committed to a target of net zero emissions by 2050, in line with the Paris Agreement that limits global warming to 1.5 degrees Celsius (Taneja, 2023). On 7 October 2022, the 184 International Civil Aviation Organization (ICAO) Member States adopted a collective long-term global aspirational goal (LTAG) of net-zero carbon emissions by 2050 (International Civil Aviation Organization, 2022). This important decision by the ICAO member states aligns with both the objectives of the Paris Agreement and the net zero carbon dioxide (CO2) emissions by the 2050 resolution agreed by the International Air Transport Association (IATA) member airlines at the 77th IATA Annual General Meeting in October 2021 (International Air Transport Association, 2022c).

With the growing focus on the impact of climate change, the embodied carbon dioxide (CO_2) emission or "carbon footprint" is now frequently used as an environmental performance indicator for products or production activities (Laurent et al., 2010). One such airline that has sustainably managed its carbon footprint is Singapore Airlines. Singapore Airlines has historically placed a very high focus on sustainable environmental management, and on managing the airline's carbon footprint. The objective of this study is to analyze Singapore Airlines annual carbon footprint from its flights, ground-based facilities energy consumption, and ground operations fuel consumption and electricity consumption. An additional objective of the study is to examine the airline's strategies to mitigate its carbon footprint as part of its commitment to environmentally sustainable operations. The study period is from 2010/11 to 2021/22 inclusive.

The remainder of the paper is organized as follows: The literature review that sets the context of the case study is presented in Section 2. The research method that underpinned the study is presented in Section 3. The Singapore Airlines case study is presented in Section 4. Section 5 presents the findings of the study.

II. BACKGROUND

2.1. Carbon Dioxide (CO_2): A Background Note

Carbon dioxide (CO_2) is one of the main greenhouse gases present in the atmosphere (Köne & Büke, 2010; Marjanović et al., 2016; Ravanchi et al., 2011). Carbon dioxide (CO_2) is naturally present in the atmosphere and is a heavy, colorless gas, which is part of the ambient air. Carbon dioxide (CO_2) is produced from the respiration and decomposition of organic substances, and from the combustion of fossil fuels and biomass (Allaby & Park, 2007). Carbon dioxide (CO_2) is the principal greenhouse gas emitted from human activities. However, the primary human activity that produces carbon dioxide (CO_2) emissions is the combustion of fossil fuels (for example, coal, natural gas, and oil) for energy and transportation. Carbon dioxide (CO_2) is also produced from some other industrial processes and land-use changes (United States Environmental Protection Agency, 2022). Carbon dioxide emissions (CO_2) means the release of greenhouse gases and/or their precursors into the atmosphere over a specified area and for a given time (Organization for Economic Cooperation and Development, 2013).

In the context of the global airline industry, an airline's carbon dioxide (CO_2) emission profile is dependent on a range of factors including, but not limited too, the aircraft deployed, commercial payload, aircraft route assignment, as well as the weather conditions for the flight (Yin et al., 2015).

2.2. Carbon Footprints

Climate change and carbon footprints are now viewed as key issues of corporate responsibility (Hrasky, 2012). According to Wiedemann and Minx (2007, p. 5), "the carbon footprint is a measure of the exclusive total amount of carbon dioxide emissions that is directly and indirectly caused by an activity or is accumulated over the life stages of a product". A carbon footprint is also the carbon emissions that are released by activities of humans (Jha et al., 2021) and from various anthropogenic activities (Ramachandra & Shwetmala, 2012). The carbon footprint (CF) is also the amount of greenhouse gases (GHG) that are emitted into the atmosphere during a product's lifecycle (Röös et al., 2013).

Carbon footprints are now being extensively used as a measure of a firm's contribution to climate change (De Grosbois & Fennell, 2011). A carbon footprint enables an organization to estimate their own contributions to the changing climate (Gautam et al., 2012). Carbon foot printing has become one of the principal methods available to firms for quantifying their anthropogenic environmental impacts and for assisting them to tackle the threat of climate change (Williams et al., 2012). The carbon footprint includes both direct and indirect emissions (Cadarso et al., 2016; Lee, 2011; Sharp et al., 2016). Thus, a carbon footprint can be segregated into direct emissions which are the greenhouse effect caused by various greenhouse gas (GHG) emissions and leakage in each process, and the indirect emissions which are the carbon dioxide (CO_2) equivalent emissions due to the energy consumption in each process (Wu et al., 2013).

2.3. The Sources of Airline Emissions

Aviation fuel is the principal source of energy consumed by the aviation industry, so both the quality and quantity of fuel affect an aircraft's carbon dioxide (CO_2) emissions (Hu et al., 2022). By consuming fuel, aircraft produce emissions of carbon dioxide (CO_2), as well as nitrogen oxides (NO_X), particles (principally soot) of sulphur oxides (SO_X), carbon monoxide (CO), as well as various hydrocarbons. First, and generating the largest percentage share, are the emissions of carbon dioxide (CO_2) which are produced in direct proportion to the volume of jet fuel used to operate flights over any distance (Sales, 2016). Water vapor is formed from the burning of jet fuels. At altitude, condensation trails from the aircraft. These comprise frozen ice crystals which deflect a small amount of sunlight away from the earth's surface and reflect more infrared radiation back toward earth. This produces an overall warming effect on the earth's atmosphere (Sales, 2013). After water vapor, carbon dioxide (CO_2) is regarded as the second most important of all the greenhouse gases (Chavan, 2016; Drewer et al., 2018; Ngo & Natowitz, 2016). The amount of aircraft carbon dioxide (CO_2) emissions is a product of hydrocarbon combustion, and the amount of these gases emitted is directly related to the volume of fuel consumed. This, in turn, is a function of the aircraft and its engine fuel efficiency (Cokorilo & Tomic, 2019), as well as the length of time that an aircraft's engines or auxiliary power unit (APU) are running (Marais et al., 2016). Carbon dioxide (CO_2) emissions are directly related to jet fuel consumption, with each kilogram of fuel consumed by aircraft producing more than three kilograms of carbon dioxide (CO_2) (Mrazova, 2014). Carbon dioxide (CO_2) are now regarded as being the most important aircraft emissions (Postorino & Mantecchini, 2014). In addition, aircraft taxi-out procedures, which form part of the aircraft Landing and Take-Off (LTO) cycle, generate substantial amounts of carbon emissions (Postorino et al., 2019). As previously noted, another source of emissions come from the aircraft auxiliary power units (APU). Because jet fuel is utilized as the power source for APUs, they emit exhaust gases (Culberson, 2011). APU's supply the essential power requirements for an aircraft whilst it is on the ground in between flights (Ashford et al., 2013; Kazda & Caves, 2015). Aircraft parked at the airport gates during their turnaround require power and air conditioning, which is typically provided by fossil fuel-combusting equipment (Greer et al., 2021), all of which produce harmful emissions.

Ground support equipment (GSE) also produces exhaust emissions. Ground service equipment (GSE) refers to vehicles and equipment that are used in the airport precinct to service aircraft whilst they are at the gate in between flights (Hazel et al., 2011). Waste disposal, aircraft de-icing usage, and wastewater processing also contribute emissions at an airport (Sahinkaya & Babuna, 2021). Other emissions are produced from aircraft maintenance and cleaning processes at an airport (Graham, 2018).

2.4. The Greenhouse Gas Protocol

The Greenhouse Gas Protocol has established a comprehensive global standardized framework to measure and manage greenhouse gas (GHG) emissions from both the private and public sectors, through value chains, and mitigation actions (Greenhouse Gas Protocol, 2022). The Greenhouse Gas Protocol categorizes greenhouse gases into both direct and indirect emissions and further categorizes them into Scope 1, Scope 2, and Scope 3 emissions (Jones, 2009). Scope 1, direct emissions, includes those emissions from sources that are owned or controlled by the firm (Girella, 2018; Vásquez et al., 2015). Scope 2, indirect emissions, come from the purchase of electricity, heat, steam or cooling. Scope 3 emissions are all the other indirect emissions that arise from the consequences of the various activities undertaken by a firm but occur from sources that are not owned nor controlled by that firm (Mazhar et al., 2019).

Although there are variations in air quality regulations by country (Budd, 2017; Elsom, 2013), airlines are now increasingly recording and reporting emissions in terms of Scope 1, Scope 2, and, in some cases, Scope 3 emissions (Baxter, 2021b).

2.5. Carbon Offsetting and Reduction Scheme for International Aviation (CORSIA)

At the 2015 Paris Climate Conference or COP 21, a landmark agreement that dealt with the adverse impact of climate change was agreed between governments (Fischlin & Ivanova, 2017; Rhodes, 2016; Viñuales et al., 2017). This significant step provided a very positive motivation for the air transport industry to develop a global market-based measure (MBM) to reduce carbon dioxide (CO_2) emissions (Singapore Airlines, 2017). In October 2016, the Member States of the International Civil Aviation Organization (ICAO) reached an agreement to adopt a global market-based measure for aviation emissions (Attanasio, 2018).

In 2021, an increasing share of the carbon emission growth in international air transport was subject to offsetting under the ICAO "Carbon Offsetting and Reduction Scheme for International Aviation" (CORSIA) program (Maertens et al., 2019). The International Air Transport Organization "CORSIA" program is a worldwide based market-based measure that has been designed to offset international aviation carbon dioxide (CO_2) emissions. The "CORSIA" program aims to stabilize the levels of such emissions from 2020 onwards. The offsetting of carbon dioxide (CO_2) emissions in the air transport industry will be achieved through the acquisition and cancelation of emissions units from the global carbon market by aircraft operators (Federal Aviation Administration, 2022). The "CORSIA" program will be rolled out in three phases with the pilot phase operating from 2021-2023. The first phase will be from 2024 to 2026. Both the pilot and first phases are voluntary. The second phase of the program is targeted at the 2027 to 2035 time period (Javed et al., 2019). Following the pilot and first phase, a second mandatory scheme will enter in effect for all ICAO Member States, except for some least developed countries (Scott &Trimarchi, 2020).

2.6. Airline Passenger Carbon Offset Programs

Carbon offsetting has become an integral element of the airline industry strategy to reduce its carbon emissions (Becken & Mackey, 2017). Consequently, airlines are now offering carbon offset schemes for their passengers so that they can reduce their carbon footprint from their travel (Chen, 2013; Ritchie et al., 2020; Zhang et al., 2019). Voluntary carbon offsetting by airline passengers is a useful measure that could help reduce environmental damage caused by air travel (Babakhani et al., 2017). In the global airline industry, carbon offsetting is a means for individuals or firms, in this case airline passengers and corporate customers, to "neutralize" their proportion of an aircraft's carbon emissions on a particular journey through an investment in carbon reduction projects (International Air Transport Association, 2022a). The principle of carbon offsetting is that the emissions for each flight are allocated amongst the passengers. Each passenger can therefore pay to offset the emissions caused by their portion of the flight's emissions. Passengers can offset their emissions through an investment in carbon reduction projects that generate carbon credits (International Air Transport Association, 2016).

Passengers participating in carbon offset programs can purchase carbon credits generated by certified renewable energy and energy efficiency projects in developing countries. These projects have been verified that they will reduce greenhouse gas emissions. A carbon credit is a permit that represents one tonne of carbon dioxide (CO_2)

that has either been removed from the atmosphere or alternatively saved from being emitted. Once used these carbon credits are subsequently "cancelled" on an official register to ensure that they cannot be sold or used again (International Air Transport Association, 2016).

Carbon credits establish a market for the reduction in greenhouse emissions by providing a monetary value to the cost of polluting the air (International Air Transport Association, 2016).

III. RESEARCH METHODOLOGY

3.1 Research Method

This study used a qualitative longitudinal research design (Derrington, 2019; Hassett & Paavilainen-Mäntymäki, 2013; Neale, 2018). Qualitative longitudinal research aims to expand and develop theories (Derrington, 2019). The researcher's role when conducting case study research is to expand and generalize theories (analytical generalization) (Rahim & Baksh, 2003). Qualitative longitudinal research aims to expand and develop theories (Derrington, 2019). A case study enables the researcher(s) to explore complex phenomena (Remenyi et al., 2010; Taber, 2014; Yin, 2018). Case studies also enable the researcher(s) to collect rich, explanatory information that provides in-depth insights into the phenomenon under investigation (Ang, 2014).

3.2 Data Collection

The data used in the study was obtained from a range of documents, company materials available on the internet and records as sources of case evidence. Documents included the Singapore Airlines annual sustainability reports, and the airline's websites. An extensive search of the leading air transport journals and magazines was also conducted in the study.

The key words used in the database searches included "Singapore Airlines sustainability policy", "Singapore Airlines climate pledge", "Singapore Airlines passenger carbon offset program", "Singapore Airlines membership of CORSIA", "Singapore Airlines annual Scope 1 carbon dioxide emissions (CO_2) from passenger and freighter aircraft operations and ground operations", "Singapore Airlines annual Scope 2 carbon dioxide emissions from electricity consumption (CO_2)", "Singapore Airlines annual carbon dioxide (CO_2) intensity ratios", and "Singapore Airlines carbon dioxide (CO_2) reduction measures".

This study used secondary data. The three principles of data collection as suggested by Yin (2018) were followed: the use of multiple sources of case evidence, creation of a database on the subject and the establishment of a chain of evidence.

3.3 Data Analysis

The data collected for the case study was examined using document analysis. Document analysis is quite commonly used in case studies. Document analysis focuses on the information and data from formal documents and a firm's records that are collected by a researcher(s) when conducting their case study (Andrew et al., 2011; Yin, 2018). Following the recommendations of Scott (2004, 2014) and Scott and Marshall (2009), the study's documents were examined according to four criteria: authenticity, credibility, representativeness and meaning.

The document analysis was undertaken in six distinct stages:

- Phase 1: The first phase involved planning the types and required documentation and their availability for the study.
- Phase 2: The data collection phase involved sourcing the documents and developing and implementing a scheme for the document management.
- Phase 3: The collected documents were examined to assess their authenticity, credibility and to identify any potential bias.
- Phase 4: The content of the collected documents was carefully examined, and the key themes and issues were identified.
- Phase 5: This phase involved the deliberation and refinement to identify any difficulties associated with the documents, reviewing sources, as well as exploring the documents content.
- Phase 6: In this phase the analysis of the data was completed (O'Leary, 2004, p. 179).

Following the guidance of Yin (2018), the study's documents were downloaded and stored in a case study database. All the documents gathered for the study were all written in English. Each document was carefully read, and key themes were coded and recorded in the case study research framework (Baxter, 2021a).

IV. RESULTS

4.1. A Brief Overview of Singapore Airlines

Singapore Airlines was established on 28 January 1972. The establishment of the new airline followed the emergence of Singapore as a Republic. Singapore had become independent from the Federation of Malaysia in 1965 (Trocki, 2006). As a result, Malaysia-Singapore

Airlines (MSA) was split into two individual airlines that subsequently became the national flag carriers of Singapore and Malaysia, respectively (Chant, 1997; Huat, 2016). Singapore Airlines commenced commercial operations on 1 October 1972 (Chan, 2000; Green & Swanborough, 1975; Seth, 2006). Singapore Airlines began serving the same international destinations that had been previously served by Malaysia-Singapore Airlines (MSA). During this period, Singapore Airline operated a fleet of Boeing B707 and Boeing B737 aircraft on these services (Chant, 1997).

On 2 April 1973, Singapore Airlines commenced daily services between Singapore and London. On 31 July 1973, the airline commenced a major expansion program following the delivery of its first wide body aircraft type, the four-turbofan powered Boeing B747-212B aircraft. This was followed quite soon thereafter by another wide body aircraft, the McDonnell-Douglas DC10-30. These aircraft were operated on the airline's medium-and-high density air routes (Chant, 1997). In the late 1970s, Singapore Airlines began operating services across the Pacific, terminating at San Francisco (Brimson, 1985).

On 20 December 1980, Singapore Airlines received its first Airbus A300B4-203 aircraft. Following its introduction into commercial service, this aircraft type complemented the Boeing B747-212B and McDonnell Douglas DC10-30 aircraft (Chant, 1997). In June 1994, Singapore Airlines announced an order for Boeing 747s, Airbus A340, plus the lease of eight Boeing 777 aircraft (Birtles, 1998). As of September 1997, Singapore Airlines had placed firm orders for thirty - four Boeing 777s (Upton, 1998). Singapore Airlines was the first airline to place an order for the Airbus A380 aircraft (Jackson, 2021; Materna et al., 2015). Singapore Airlines subsequently took delivery of its first Airbus A380 aircraft on the 15th of October 2007. Singapore Airlines was the first airline in the world to operate the Airbus A380 aircraft. On October 15, 2007, the first Airbus A380-800 aircraft was delivered to Singapore Airlines. Singapore Airlines began Airbus A380 commercial operations on 25 October 2007, when it commenced daily Airbus A380 flights between Singapore and Sydney (Kingsley-Jones, 2008; Simons, 2014). Singapore Airlines was the launch customer for Boeing Commercial Airplanes Boeing 787-10 aircraft (Polek, 2017).

At the time of the present study, Singapore Airlines operated a modern passenger fleet of 123 aircraft and had outstanding orders for a further 44 aircraft. SIA Cargo operated a fleet of 7 Boeing B747-400 freighter aircraft (Singapore Airlines, 2022). The Singapore Airlines Group comprises the wholly owned subsidiaries Scoot Airways, and SIA Cargo. On 25 July 2017, Scoot and Tigerair, the SIA Group low-cost carriers, were officially merged into a single entity that retained the Scoot brand (Gupta Kapoor, 2017; Ong, 2017; Singapore Airlines, 2019). Singapore Airlines formally joined the Star global passenger alliance in 2000 (Heracleous & Wirtz, 2012; Iatrou & Alamdari, 2005; Ramaswamy, 2002). As at September 2021, Silkair had been fully integrated into Singapore Airlines (Singapore Airlines, 2022).

Singapore Airlines has adopted the full-service network carrier business model. According to Ehmer et al. (2008, p. 5), a "full-service network carrier is an airline that focuses on providing a wide range of pre-flight and onboard services, including different service classes, and connecting flights". As of March 31, 2022, the airline's combined passenger network covered 69 destinations (Singapore Airlines, 2022).

Figure 1 presents Singapore Airlines annual enplaned passengers and revenue passenger kilometres performed (RPKs) for the period covering the financial years 2010/2011 to 2021/2022. One passenger enplanement measures the embarkation of a revenue passenger, whether originating, stop-over, connecting or returning (Holloway, 2016). One revenue passenger kilometre (RPK) is one passenger transported one kilometre (Belobaba, 2016; Gillen, 2017). Over the period FY2010/2011 to 2019/2020, Singapore Airlines enplaned passengers and RPKs showed quite consistent growth (Figure 1). Figure 1 shows, however, that there was a very steep decline in both the annual number of enplaned passengers and RPKs in 2020, which were both adversely impacted by the global COVID-19 pandemic. In 2020, the COVID-19 pandemic caused a decline in economic activity around the world. This decline in economic activity caused a major disruption in the air travel market supply and demand chain (Dube et al., 2021). Air transportation was one of the sectors most adversely impacted by the corona-virus pandemic (Barczak et al., 2022; Karakuş et al., 2021; Sun et al., 2022). In addition, due to the global coronavirus crisis, most countries placed restrictive measures to confine the pandemia (Maria Iacus et al., 2020). As a result, these restrictions had a very adverse impact on global airline passenger demand, and thus, on the total number of revenue passenger kilometres performed (RPKs) by the world's airlines in 2020 (International Air Transport Association, 2021). On 24th March 2020, the Singaporean Government closed its international borders (Djalante et al., 2020). Figure 1 shows that Singapore Airlines annual enplaned passengers grew from 457,000 in the 2020/2021 financial year to 3,388,000 enplaned passengers in the 2021/2022 financial year. In the 2021/2022 financial year, the airline's revenue passengers' kilometres performed (RPKs) increased by 642.75% on the

2020/2021 financial year levels. The increase in both enplaned passengers and RPKs in the 2021/2022 financial year, reflected the growth in the airline's operations as COVID-19 pandemic measures were eased throughout its route network.

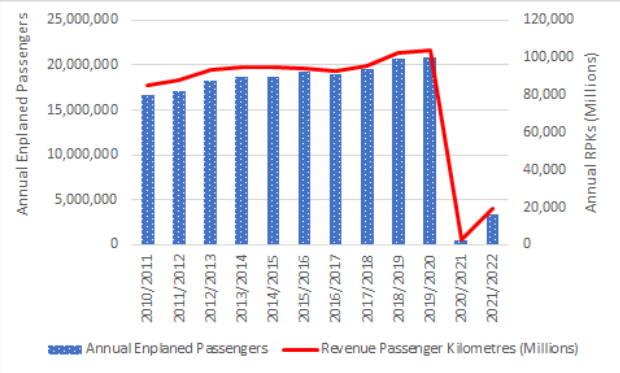

Fig.1: Singapore Airlines annual enplaned passengers and revenue passenger kilometres performed (RPKs): 2010/11-2021/22. Note: Financial years from 1 April to 31 March. Source: Data derived from Singapore Airlines (2013, 2016, 2021, 2022).

4.2. Singapore Airlines Environmental Policy

Singapore Airlines has implemented a very comprehensive environment and sustainability related policy that is underpinned by four key pillars.

Pillar One: Improved technology: Singapore remains committed to its long-held policy of operating a modern and fuel-efficient fleet (Singapore Airlines, 2019). Singapore Airlines took delivery of its first Boeing 787-10 aircraft on March 25, 2018 (Australian Aviation, 2018; Singapore Airlines, 2018a). On September 22, 2018, Airbus delivered the first A350-900 Ultra Long Range (ULR) aircraft to Singapore Airlines (Airbus, 2018). As of 31 March 2022, Singapore Airlines operated a fleet of 123 aircraft, with an average age of five years and seven months. In addition, the airline had 7 Airbus A350-900XWB, 31 Boeing 777-9s, 13 Boeing 787-10s, and 24 Boeing 737-8 MAX aircraft on firm order (Singapore Airlines, 2022). As part of its environmental policy, Singapore Airlines makes investments in engineering improvement packages for its aircraft airframes and engines. These improvements help to reduce drag and whilst at the same time improve engine efficiency, and thus, enhance fuel efficiency (Singapore Airlines, 2021).

Pillar Two: Operational measures: Importantly, as previously noted, Singapore Airlines maintains a modern and fuel-efficient fleet. As of 31 March 2022, the airline was operating 58 Airbus A350-900XWB aircraft. The Airbus A350-900XWB aircraft is acknowledged for its improved operating efficiency (Singapore Airlines, 2019). At the time of the present study, Singapore Airlines operated a fleet of fifteen Boeing 787-10 aircraft (Singapore Airlines, 2022). The Boeing 787-10 aircraft burns 20% less fuel than its predecessor the Boeing B767 (Vasigh et al., 2012). A key focus of the airline's environmental policy is on improving aircraft fuel productivity through the implementation of "green" operations and by reducing fuel usage through highly efficient aircraft weight management together with the optimization of flight routes (Singapore Airlines, 2021).

Pillar Three: Improved infrastructure: Infrastructure improvements in the air transport industry present an opportunity for airlines to reduce both fuel use and carbon dioxide (CO_2) emissions. Such improvements can be achieved from the optimization of air routes through efficient air traffic control management. Singapore Airlines collaborates with Air Traffic Management stakeholders to investigate new ways to improve and enhance airspace incremental efficiency (Singapore Airlines, 2019, 2021).

Pillar Four: Global Market-Based Measure (MBM): Singapore Airlines recognizes that the International Civil Aviation Organization (ICAO) CORSIA scheme will play an essential role in achieving carbon neutral growth in a cost-efficient manner. As such, Singapore Airlines fully supports the ongoing efforts towards meeting the requirements in the ICAO "CORSIA" Monitoring, Reporting and Verification system of carbon emissions. As of 1 January 2019, Singapore Airlines commenced the monitoring and reporting of its carbon dioxide (CO_2) emissions on an annual basis. Furthermore, Singapore Airlines will voluntarily participate in the ICAO CORSIA program from 2021 to 2026, following which the scheme becomes mandatory (Singapore Airlines, 2019).

The International Civil Aviation Organization (ICAO) is responsible for setting the international standards for nitrous oxides (NOx), carbon dioxide (CO2), hydrocarbons, and exhaust levels from aircraft engine emissions in accordance with Annex 16 (Volume III) to the 1944 Chicago *Convention on International Civil Aviation* (International Civil Aviation Organization, 2014; Young, 2018). As part of its sustainable environment policy, Singapore Airlines fully supports the new carbon dioxide (CO_2) emissions standard that has been set by the International Civil Aviation Organization (ICAO) in order meet the air transport industry's long-term commitment to reduce carbon dioxide (CO_2) emissions (Singapore Airlines, 2021).

Singapore Airlines is dedicated to its long-term responsibility to protect the environment while delivering air transportation services that are of the highest quality.

As discussed below, Singapore Airlines has introduced an extensive range of programs to enable it to implement sustainable practices across its operations in a responsible manner. The airline actively manages issues such as carbon dioxide (CO_2) emissions, noise, waste, as well as energy and water consumption. Singapore Airlines continues to explore new sustainable practices across all areas of its operations. The company has adopted the International Air Transport Association (IATA) four-pillar strategy to address climate change, and, as a result, it seeks opportunities to reduce the carbon footprint of its operations. The airline also promotes eco-friendly habits among its employees and stakeholders. In addition, Singapore Airlines raises the awareness on the importance of taking action to reduce its impact on the environment (Singapore Airlines, 2020). In 2021, the Singapore Airlines Group (SIA) committed to a net zero carbon emissions standard across all operations by 2050 (Asian Aviation, 2021; Becken, 2021). According to the Climate Council (2022), "net zero emissions' refers to achieving an overall balance between greenhouse gas emissions produced and greenhouse gas emissions taken out of the atmosphere".

Singapore Airlines became a signatory to the Ten Principles of the United Nations Global Compact in 2018 (Singapore Airlines, 2019). Singapore Airlines has prioritized three of the seventeen United Nations (UN) Development goals, that is, Goal Number 8 (Promote sustained, inclusive, and sustainable economic growth, full and productive employment, and decent work for all), Goal 12 (Ensure sustainable consumption and production patterns), and Goal 13 (Take urgent action to combat climate change and its impact), The airline has also adopted UN Development Goal 7 affordable and clean energy (Singapore Airlines, 2021). In 2015, all United Nations Member States adopted the "2030 Agenda for Sustainable Development" and its seventeen 17 Sustainable Development Goals (SDGs). Each SDG comprises a range of targets to be achieved by 2030 (Katila et al., 2019; United Nations, 2022). The United Nations Sustainable Development Goals (SDGs) provide a framework for business and government to solve global economic, social, and environmental challenges (Air New Zealand, 2018).

On 10 February 2021, the Singaporean Government announced its "Singapore Green Plan 2030", that charts the country's targets for sustainability for the forthcoming decade (Chng & Ong, 2021; Lin-Heng, 2021). In line with the "Singapore Green Plan 2030", as well as Singapore Airlines Sustainability Policy and Climate Action Pledge, the airline is continuing to explore and implement new sustainable practices across its operations. These include a focus on the use of green energy, green infrastructure, and buildings, as well as green citizenry, which involves reducing water consumption, and reducing waste disposed to landfill (Singapore Airlines, 2022).

4.3. Singapore Airlines Annual Scope 1 Carbon Dioxide (CO_2) Emissions from Flight and Ground Operations

Direct Scope 1 greenhouse gas (GHG) emissions occur from sources that are owned or controlled by Singapore Airlines. These direct Scope 1 emissions include emissions from aircraft jet fuel and fuel consumption in Singapore Airlines owned or controlled vehicles (Singapore Airlines, 2020).

4.3.1. Annual Carbon Dioxide (CO_2) Emissions from Freighter Services

The provision of air freight services is a core business product of Singapore Airlines. As previously noted, SIA Cargo, the air cargo division of Singapore Airlines, operates a fleet of 7 Boeing B747-400 freighter aircraft. A freighter aircraft is an aircraft that has been expressly designed or which has been converted to transport air cargo, express, and so forth, rather than passengers (Wensveen, 2016). The Boeing B747-400 freighter is 10-16 per cent more fuel efficient than its 747-200 model (Wallis, 2013), and this additional fuel efficiency translates into lower aircraft emissions. Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from the operation of SIA Cargo freighter services and the year-on-year change (%) from the 2010/11 financial year to the 2021/22 financial year is presented in Figure 2. As can be observed in Figure 2, SIA Cargo annual freighter aircraft carbon dioxide (CO_2) emission levels have predominantly displayed a downward trend, decreasing from a high of 1,716.87 thousand tonnes in the 2010/11 financial year to a low of 874.7 thousand tonnes in the 2018/19 financial year. This downward trend is demonstrated by the year-on-year percentage change line graph, which is more negative than positive, that is, more values are below the line than above. Figure 2 shows that during the study period, there were just three years, when the SIA Cargo freighter carbon dioxide (CO_2) emissions increased on a year-on-year basis. These increases were recorded in the 2014/2015 financial year (+4.18%), the 2020/21 financial year (+17.05%), the 2021/2022 financial year (+2.36%), respectively (Figure 2). These increases could be attributed to greater freighter flight frequencies in these respective years. The largest single annual decrease in carbon dioxide (CO_2) emissions occurred in the 2012/13 financial year, when these aircraft emissions decreased by 38.68% on the previous year levels (Figure 4). The overall downward trend is highly favorable and shows that SIA Cargo has been able to accommodate the growing demand for their services while at the same time mitigating as much as possible the detrimental impact of their freighter aircraft carbon dioxide (CO_2) emissions on the environment.

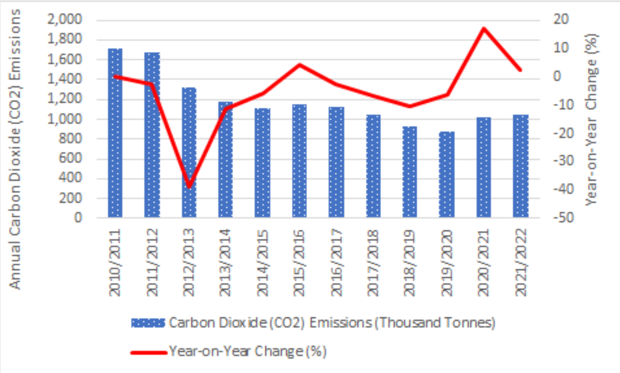

Fig.2: Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from SIA Cargo freighter services and the year-on-year change (%): 2010/11-2021/22. Note: Financial years from 1 April to 31 March.

Source: Data derived from Singapore Airlines (2013, 2014, 2017, 2022).

The SIA Cargo annual carbon dioxide (CO_2) freighter aircraft emissions intensity ratio ($KgCO_2$/LTK) and the year-on-year change from the 2010/11 financial year to the 2021/22 financial year is depicted in Figure 3. Load tonne per kilometre (LTK) is a unit of measurement for transport capacity and represents the transport of a tonne (1000 kilograms) of load capacity over one kilometre (Statistics Netherlands [CBS], 2022). Figure 3 shows that the SIA Cargo annual freighter aircraft carbon dioxide emissions intensity ratio oscillated over the study period, rising from a low of 0.53 Kg CO_2/LTK to a high of 0.58 Kg CO_2/LTK in the 2015/16 financial year and the 2019/20 financial year, respectively (Figure 3). The largest single annual increase in this metric occurred in 2015/2016 and 2019/2020 financial years, when the annual value of this ratio increased by 5.45% in both years (Figure 3). The largest single annual decrease in this ratio was recorded in the 2017/18 financial year, when there was a decrease of 3.63% on the previous year level (Figure 3). The decrease in the 2020/21 financial year (-3.44%) was attributed to lower levels of freighter flight activity associated with the downturn in demand from the Covid 19 pandemic. This ratio decreased by 1.78% in the 2021/2022 financial year, which once again was a favorable result given the increase in freighter services in the 2021/2022 financial year.

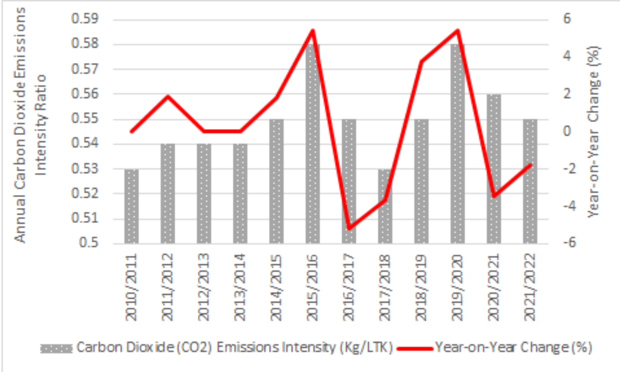

Fig.3: Singapore Airlines annual carbon dioxide (CO_2) emissions intensity ratio ($KgCO_2$/LTK) for SIA Cargo freighter services and the year-on-year change: 2010/11-2020/21.

Source: Data derived from Singapore Airlines (2013, 2014, 2017, 2022).

4.3.2. Annual Carbon Dioxide (CO_2) Emissions from Passenger Services

Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from its passenger services and the year-on-year change (%) from the 2010/11 financial year to the 2021/22 financial year is presented in Figure 4. As can be observed in Figure 4, despite the increase in the number of passengers carried and the higher number of services operated, Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from passenger services remained relatively constant over the period 2010/11 to 2019/2020. During this period, the highest single year-on-year change in emissions, was recorded in the 2012/13 financial year, at which time the annual carbon dioxide (CO_2) emissions increased by 4.97% on the previous year levels (Figure 4). Figure 4 shows that the airline's passenger service carbon dioxide (CO_2) emissions decreased on a year-on-year basis in the 2014/15 financial year, when the annual carbon dioxide (CO2) emissions decreased by 1.93% on the 2013/14 financial year levels (Figure 4). Figure 4 also shows that was a very significant decrease (-78.22%) in the airline's annual carbon dioxide (CO_2) levels in the 2020/21 financial year. This substantial decrease was due to the greatly scaled back level of operations because of the global Covid-19 pandemic (Singapore Airlines, 2021). Figure 4 shows that there was a pronounced spike in this metric during the 2021/2022 financial year, at which time it increased by 126.1% on the previous year levels. In the 2021/2022 financial year, Singapore Airlines was able to operate more passenger services following the gradual easing of COVID-19 pandemic measures throughout its route network. In the 2021/2022 financial year, Singapore Airlines increased the number of destinations that it served from 47 destinations in the 2020/2021 financial year to 69 destinations in the 2021/2022 financial year.

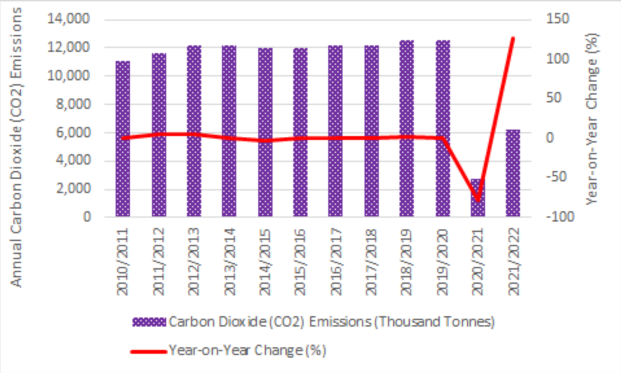

Fig.4: Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from passenger services and the year-on-year change (%): 2010/11-2021/22.

Source: Data derived from Singapore Airlines (2013, 2014, 2017, 2022).

Figure 5 presents Singapore Airlines annual passenger services carbon dioxide (CO_2) emissions intensity ratio (Kg CO_2/LTK) and the year-on-year change from the 2010/11 financial year to the 2021/22 financial year is presented in Figure 5. Figure 5 shows that there were three discrete trends with this ratio. From the 2010/11 financial year to the 2012/13 financial year, there was an upward trend with the value of the ratio increasing from 0.93 Kg CO_2/LTK to 0.95 KgCO_2/LTK in the 2012/13 financial year (Figure 5). Over the period 2012/13 to 2018/19, the ratio decreased from a high of 0.95 Kg CO_2/LTK in the 2012/13 financial year to a low of 0.88 KgCO_2/LTK in the 2018/19 financial year (Figure 5). This was a very favorable result given the increase in the number of passengers carried and the higher level of services operated by the airline. Figure 5 shows that there was a pronounced spike in this metric in the 2020/21 financial year, when it increased by 28.08% on the 2019/20 level. This increase may be due to the smaller number of passengers carried as well as the lower number of flights operated due to the COVID-19 virus pandemic. These two factors resulted in the ratio increasing from 0.89 KgCO_2/LTK in 2018/19 to a high of 1.14 KgCO_2/LTK in 2020/21 (Figure 5). Figure 5 shows that this metric decreased on a year-on-year basis in the 2021/2022 financial year by 4.38%. This was a very favorable outcome as the airline passenger traffic increased by 641.35% whilst carbon dioxide (CO_2) emissions from its passenger services increased by 126.1%. There was a further decrease in this metric in the 2021/2022 financial year, at which time it decreased by 4.38% on the previous year levels (Figure 5). This decrease suggests that Singapore Airlines was able to increase its passenger traffic in the 2021/2022, whilst at the same time decreasing the amount of carbon dioxide (CO_2) emissions per enplaned passenger.

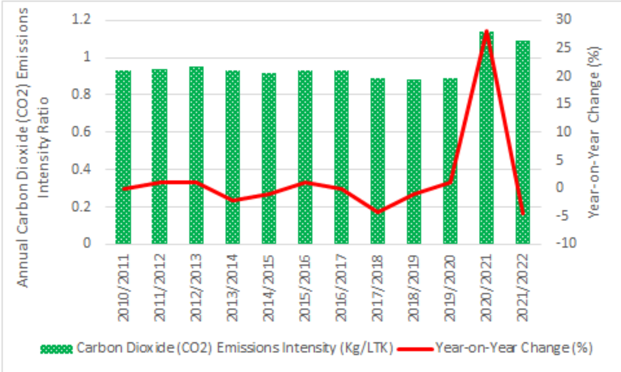

Fig.5: Singapore Airlines annual carbon dioxide (CO_2) emissions intensity ratio (KgCO_2/LTK) for passenger services and the year-on-year change: 2010/11-2020/21.

Source: Data derived from Singapore Airlines (2013, 2014, 2017, 2022).

4.3.3. Total Annual Carbon Dioxide (CO_2) Emissions from Flight Operations

Singapore Airlines total annual scope 1 carbon dioxide (CO_2) emissions from passenger and freighter operations and the year-on-year change (%) from the 2010/11 financial year to the 2020/22 financial year is presented in Figure 6. Figure 6 shows that the total annual Scope 1 carbon dioxide (CO_2) emissions from the total flight operations (passenger and freighter flights) oscillated over the study period. There was a slight annual increase in these carbon dioxide (CO_2) emissions over the financial years covering the period 2010/11 to 2012/13. Following this there was a slight downward trend in the 2013/14 financial year (-1.04%), and again in the 2014/15 financial year (-2.25%), respectively (Figure 6). The airlines total annual scope 1 carbon dioxide (CO_2) emissions from flight operations grew on a year-on-year basis from the 2015/16 financial year to the 2018/19 financial year. During this time frame, the largest single annual increase in carbon dioxide (CO_2) emissions was recorded in the 2018/19 financial year, when these emissions increased by 1.76% on the previous year's levels (Figure 6). In the latter years of the study, that is, from the 2019/20 financial year and the 2020/21 financial year, there was a downward trend in these emissions, with a very significant decrease recorded in the 2020/21 financial year (-72.03%) (Figure 6). The sharp decrease in the 2020/21 financial year was attributed to lower levels of flight activity associated with the downturn in demand from the COVID 19 pandemic. Figure 6 shows that this metric increased by 92.42% in the

2021/2022 financial year, reflecting the increase in both passenger and freighter services operated by the airline. As outlined below, Singapore Airlines has implemented a wide range of carbon dioxide (CO_2) reduction measures over the study period, and these measures have enabled the airline to mitigate its carbon footprint.

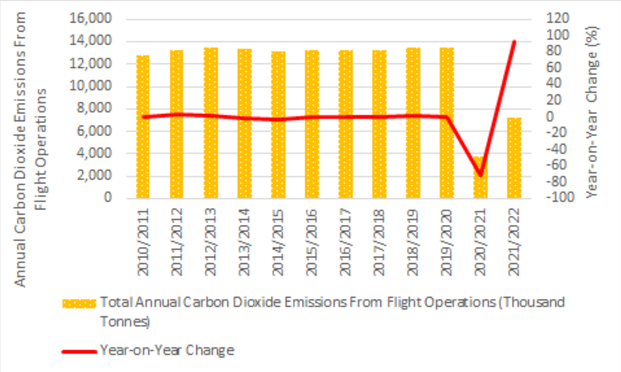

Fig.6: Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from flight operations and the year-on-year change (%): 2010/11-2021/22.

Source: Data derived from Singapore Airlines (2013, 2014, 2017, 2022).

4.3.4. Annual Carbon Dioxide (CO_2) Emissions from Ground Operations

Throughout the study period, Singapore Airlines operated a fleet of ground-based vehicles that were powered by either petrol or diesel engines. These vehicles are required to comply with the National Environmental Agency's (NEA) requirements on motor vehicle emissions (Singapore Airlines, 2021). Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from fossil fuel consumption and the year-on-year change for the period covering the 2010/11 financial year to the 2021/22 financial year is presented in Figure 7. As can be observed in Figure 7, the annual carbon dioxide (CO_2) emissions from fossil fuel consumption fluctuated over the study period. The largest single annual decrease in these emissions was recorded in 2016/2017 financial year (-99.43%) (Figure 7). The highest single annual increase in these emissions was in the 2021/2022 financial year, when they increased by 28.07% on the 2020/2021 financial year levels. Figure 7 also shows that there was a considerable decrease in these emissions in the 2014/15 financial year of -10.35% (Figure 7). In the latter years of the study period, there was a very significant decrease in the airline's annual fossil fuel related carbon dioxide (CO_2) emissions. From an environmental perspective, these carbon dioxide (CO_2) emissions have decreased significantly in the latter years of the study period, that is, from the 2016/2017 financial year to 2021/2022 financial years, and suggest that Singapore Airlines has been able to reduce its overall ground operations carbon dioxide (CO_2) emissions very effectively, and hence, mitigate the impact of these emissions upon the environment.

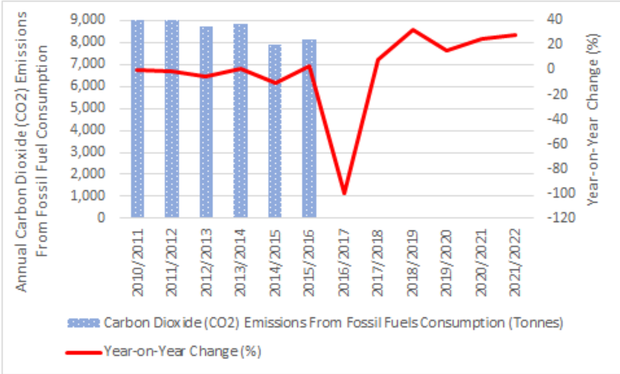

Fig.7: Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from ground operations and the year-on-year change (%): 2010/11-2021/22.

Source: Data derived from Singapore Airlines (2013, 2014, 2017, 2022).

4.3.5. Annual Carbon Dioxide (CO_2) Emissions from Diesel Consumption

Singapore Airlines indirect (Scope 2) carbon dioxide (CO_2) emissions from its ground vehicle diesel consumption and the associated year-on-year change (%) from the 2017/18 financial year to the 2021/22 financial year is presented in Figure 8. As can be observed in Figure 8, there has been an overall upward trend in this metric, when it increased from a low of 8.9 tonnes in the 2017/2018 financial year to a high of 29.2 tonnes in the 2021/2022 financial year. This overall upward is demonstrated by the year-on-year percentage change line graph, which is more positive than negative, that is, all values are above the line. Figure 8 shows that there were two very substantial spikes in this metric during the study period. These significant increases were recorded in the 2018/2019 financial year (+59.55%) and in the 2019/2020 financial year (+91.54%), respectively (Figure 8), and reflected an increased demand for diesel fuel in both financial years. Figure 8 shows that there were no recorded annual decreases in this metric during the study period.

4.3.6. Annual Carbon Dioxide (CO_2) Emissions from Petrol Consumption

Singapore Airlines indirect (Scope 1) carbon dioxide (CO_2) emissions from its ground vehicle petrol consumption and the associated year-on-year change (%) from the 2017/2018 financial year to the 2021/2022 financial year are presented in Figure 9. Figure 9 shows that the annual carbon dioxide (CO_2) emissions from its petrol consumption increased by 20.32% in the 2018/2019 financial year, reflecting a higher demand for petrol in that financial year (Figure 9). There was a pronounced decrease in this metric during the 2020/2021 financial year, at which time it decreased by 38.55% on the previous year levels (Figure 9). As previously noted, Singapore Airlines was adversely impacted by the COVID-19 pandemic and the related government pandemic response measures in 2020. These factors influenced Singapore Airlines petrol consumption in the 2020/2021 financial year, and due to the lower demand for petrol there was an associated decrease in the airline's carbon dioxide (CO_2) emissions because of the lower petrol consumption in the 2020/2021 financial year. Figure 9 shows that there was a significant spike in this metric during the 2021/2022 financial year, at which time it increased by 53.69% on the 2020/2021 financial year levels. This increase could be attributed to the increased operational activities associated with the increase in flights operated by Singapore Airlines in the 2021/2022 financial year. As noted earlier, Singapore Airlines expanded the number of destinations that it served in the 2021/2022 financial year.

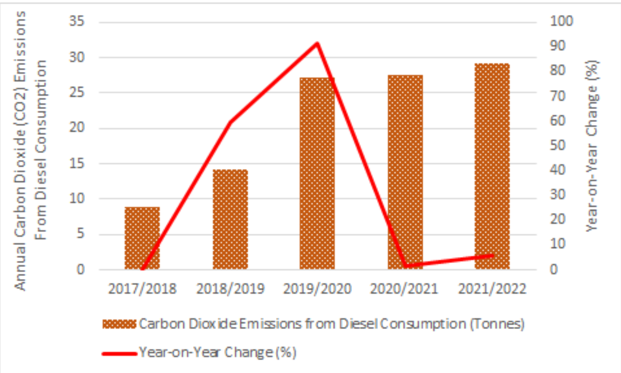

Fig.8: Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from diesel consumption and the year-on-year change (%): 2010/11-2021/22.

Note: Data prior to 2017/2018 not available.

Source: Data derived from Singapore Airlines (2013, 2017, 2022).

4.4. Singapore Airlines Annual Scope 2 Carbon Dioxide (CO_2) Emissions

Singapore Airlines indirect (Scope 2) greenhouse gas (GHG) emissions are those emissions that are produced from the generation of purchased electricity that is consumed in Singapore Airlines buildings (total gross floor area) and offices (total leased area). These emissions physically occur at the facilities where electricity is generated (Singapore Airlines, 2020). Figure 10 presents Singapore Airlines annual Scope 2 carbon dioxide (CO_2) emissions from indirect energy consumption (electricity) and the year-on-year change for the period covering the 2010/11 financial year to the 2021/2022 financial year. As can be observed in Figure 10, the annual Scope 2 carbon dioxide (CO_2) emissions from indirect energy (electricity) consumption have exhibited an overall downward trend during the study period. As discussed below, Singapore Airlines has implemented a wide range of energy efficiency measures that have played a key role in reducing the airline's annual carbon dioxide (CO_2) emissions from its electricity consumption. This overall downward trend is demonstrated by the year-on-year percentage change line graph, which is more negative than positive, that is, more values are below the line than above. Indeed, Figure 10 shows that there were only two annual increases in Singapore Airlines annual indirect Scope 2 carbon dioxide (CO_2) emissions from its electricity consumption. These annual increases were recorded in the 2010/11 and 2021/2022 financial years. In the 2010/2011 financial year, Singapore Airlines annual carbon dioxide (CO_2) emissions from its electricity consumption increased by 1.06% on the previous year levels. In the 2021/2022 financial year Singapore Airlines annual carbon dioxide (CO_2) emissions from its electricity consumption increased by 1.46% on the previous year levels, with the increase reflecting a higher demand for electricity in the 2021/2022 financial year (Figure 10). As can be observed in Figure 10, there were three quite significant annual decreases in this metric.

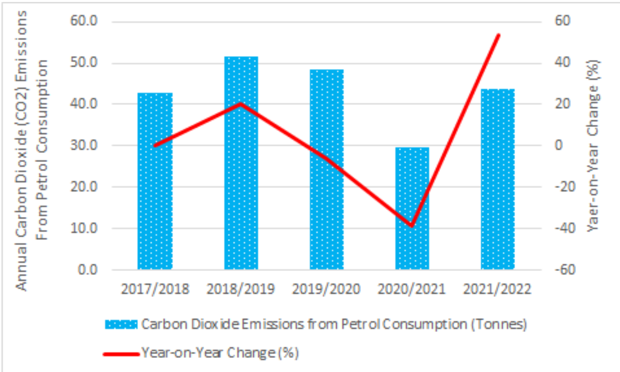

Fig.9: Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from petrol consumption and the year-on-year change (%): 2010/11-2021/22.

Note: Data prior to 2017/2018 not available.

Source: Data derived from Singapore Airlines (2013, 2016, 2021, 2022).

These decreases occurred in the 2014/2015 financial year (-10.63%), the 2017/2018 financial year (-44.03%), and the 2020/2021 financial year (-27.4%), respectively (Figure 10). In each of these years, Singapore Airlines was able to increase its passenger traffic and services, whilst at the same time reducing its annual electricity consumption, and thus, its carbon dioxide (CO_2) emissions that are produced from this energy source. This is a very favorable trend and suggests that the wide range of energy saving measures have had a significant impact on the annual level of Scope 2 carbon dioxide (CO_2) emissions from the airline's electricity consumption.

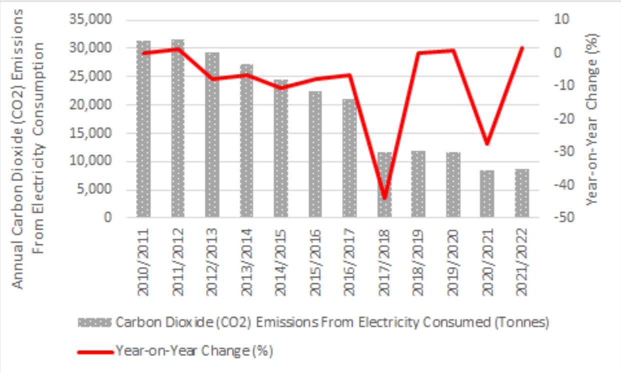

Fig.10: Singapore Airlines annual Scope 2 carbon dioxide (CO_2) emissions from electricity consumption and the year-on-year change (%): 2010/11-2021/22.

Source: Data derived from Singapore Airlines (2013, 2016, 2021, 2022).

Singapore Airlines annual Scope 2 carbon dioxide (CO_2) emissions from electricity consumption per enplaned passenger and the year-on-year change (%) for the period covering the 2010/11 to 2021/22 financial years is depicted in Figure 11. Figure 11 shows that there was an overall downward trend in this metric from the 2010/2011 financial year to the 2019/2020 financial year. During this period, there were three quite significant annual decreases in this metric. These decreases were recorded in the 2012/2013 financial year (-12.98%), the 2014/2015 financial year (-11.19%), and the 2017/2018 financial year (-45.48%), respectively (Figure 11). This was a very favorable outcome as it showed that the airline could handle higher levels of demand for its passenger services whilst at the same time reducing its electricity consumption, and hence, its carbon dioxide (CO_2) emissions per enplaned passenger. However, there was one marked exception to this trend, which occurred in the 2020/2021 financial year. Figure 11 shows that there was a very substantial spike in this metric in the 2020/2021, when it increased by 3,221.1% on the 2019/2020 financial year levels. This substantial increase could be attributed to the annual decrease in passengers (-97.81%) being a lot greater than the annual decrease in the airline's electricity consumption (-27.4%). As a result, there were fewer passengers to spread the electricity consumption related carbon dioxide (CO_2) emissions over in the 2020/2021 financial year. Figure 11 also shows that there was quite a significant decrease in this metric during the 2021/2022 financial year, when it decreased by 86.31% on the 2020/2021 financial year levels. This was another favorable outcome as it demonstrated that Singapore Airlines was once again able to handle more passengers whilst at the same time reducing its electricity consumption per enplaned passenger, and this in turn, resulted in the significant decrease in this metric in the 2021/2022 financial year.

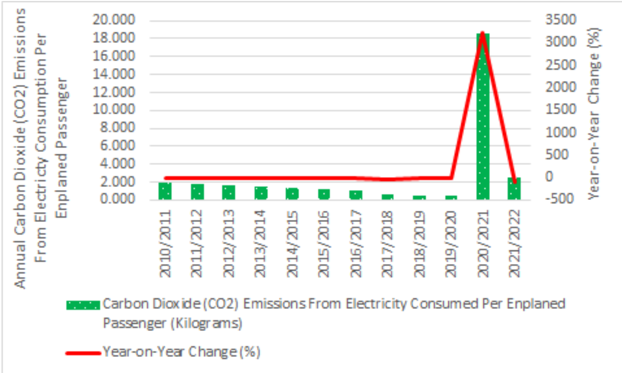

Fig.11: Singapore Airlines annual Scope 2 carbon dioxide (CO_2) emissions from electricity consumption per enplaned passenger and the year-on-year change (%): 2010/11-2021/22.

Source: Data derived from Singapore Airlines (2013, 2016, 2021, 2022).

4.5. Singapore Airlines Carbon Footprint Mitigation Measures

4.5.1. Aircraft Weight Reduction Program

A significant development in the airline industry in recent times has been the efforts made by airlines to reduce the weight of their aircraft, and thus, reduce aircraft fuel burn and the associated harmful emissions (Gilani & Körpe, 2019).

The aircraft operated by airlines on their commercial services have a potable water uplift capability which is used to service the aircraft cabin and its associated amenities. Depending upon the size of the aircraft assigned to a flight, the potable water uplift can exceed 2,000 litres (Franzi, 2018). Airlines have also implemented potable water strategies whereby they carefully optimize the water uplift on flights to satisfy passenger requirements whilst at the same time achieving aircraft fuel burn reductions and the related emissions savings from the lower aircraft weight (Baxter, 2016).

Throughout the study period, Singapore Airlines has implemented initiatives both within the airlines that are part of the Singapore Airlines Group and with the Original Equipment Manufacturers (OEMs) that are designed to reduce the weight of its aircraft fleet. An example of such a weight saving initiative was the optimization of the water uplift based on flight sector requirements through a tailored potable water program. A further weight saving initiative involved the removal of unutilized overhead storage compartments located in between the galleys of the airline's Boeing 777-300ER aircraft. These two aircraft weight initiatives saved 2,400 tonnes of fuel per year whilst also reducing carbon dioxide (CO_2) emissions by around 7,560 tonnes per annum (Singapore Airlines, 2017).

4.5.2. ASPIRE Program

The Asia and Pacific Initiative to Reduce Emissions (ASPIRE) program is a partnership of air traffic control service providers that is focused on environmental stewardship within the Asia/Pacific region (Singapore Airlines, 2014). On February 18, 2008, the ASPIRE agreement was signed in Singapore, with the original members being Australia's and New Zealand's air traffic service providers and the United States Federal Aviation Administration (FAA). The agreement was subsequently signed by the Japan, Singapore, and Thailand air traffic service providers (Nakamura et al., 2014; Woon Lee et al., 2018).

As part of the "ASPIRE-Daily City Pair' program", regular "green" flights have been conducted between pairs of airports located throughout the Asia-Pacific region. Under this program, the aviation authorities in Singapore, the United States, and Japan have also worked in partnership to ensure optimal air traffic conditions (Singapore Airlines, 2014). The first multi-sector demonstration "green flight" operated by Singapore Airlines under the ASPIRE program was the airline's flight SQ11. This flight operated from Los Angeles to Singapore, with a transit stopover in Tokyo, Tokyo on 31 January 2010. In total, the airline achieved a fuel saving of 6% as compared to a similar flight. This fuel saving translated into a reduction of 33,769 kg of carbon dioxide (CO_2) emissions. On 16 May 2011, the first scheduled commercial service between Los Angeles and Singapore, flight number SQ37, was launched as part of the program. The non-stop flight employed enhanced gate-to-gate air traffic management operational procedures that were designed to reduce both aircraft fuel burn and carbon dioxide (CO_2) emissions. Other flight optimization measures included Dynamic Airborne Reroute Procedures, 30/30 Reduced Oceanic Separation, and Time-Based Arrival Management and Arrival Optimization. These measures significantly reduce fuel burn and the carbon dioxide (CO_2) emissions on the airline's flights. According to Singapore Airlines, there is an average carbon dioxide (CO_2) emission saving of 3.8 tonnes for each Los Angeles-Singapore flight operated by the airline (Singapore Airlines, 2013). The Oceania region has been included in the ASPIRE program since 15 September 2012 (Singapore Airlines, 2015).

Under the ASPIRE program, Singapore Airlines commenced Singapore-Canberra-Wellington and vice versa flights in 2017 that used the range of ASPIRE practices. These ASPIRE practices included the use of favorable winds, reducing airborne holding, and enabling efficient continuous descent arrivals and reducing aircraft taxiing times. On an annual basis, the use of the ASPIRE Program practices would enable Singapore Airlines to reduce its aircraft fuel consumption by an estimated 1,500 tonnes of fuel and this would translate into annual saving of 4,600 tonnes of carbon dioxide (CO_2) emissions on these flight sectors (Singapore Airlines, 2017). Singapore Airline's non-stop service between Singapore and San Francisco was included in the ASPIRE program in the 2017/18 financial year (Singapore Airlines, 2018).

4.5.3. Cleaner-energy vehicles

In addition to the emissions produced from aircraft, the operation of the aircraft ground service equipment used to service aircraft during the time they are on the ground in between flights can also produce significant amounts of carbon dioxide (CO_2) emissions (International Airport Review, 2010). Thus, the replacement of internal combustion engine powered airport ground support vehicles and equipment used by airlines and ground handling agents could potentially reduce carbon dioxide (CO_2), carbon monoxide (CO), hydrocarbons (HC), nitrogen oxides (NOx), and particulate matter (PM) (Gellings, 2011). Singapore Airlines has been cognizant of the environmental impact of fossil fuel vehicles and equipment and, as a result, in the 2020/21 financial year, Singapore Airlines installed six electric vehicles (EV) charging stations and promoted the use of electric vehicles (Singapore Airlines, 2021).

4.5.4. Energy Conservation Measures

An important development in the global airline industry has been the strategy implemented by airlines to actively promote energy-saving and carbon reduction measures. These measures are being implemented to enable airlines to to reduce the greenhouse (GHG) gases that are generated in the provision of transportation services (Tsai et al., 2014). The associated environmental-related benefit of reduced energy consumption is the reduction in carbon dioxide (CO_2) emissions. Indeed, energy efficiency provides several important environmental benefits. Energy efficiency measures can reduce greenhouse (GHG) emissions, both direct emissions from fossil fuel combustion and consumption, as well as the indirect emissions reductions from electricity generation (International Energy Agency, 2022).

Throughout the study period, Singapore Airlines has introduced a range of facility and building-related energy saving measures, which had a particular focus on air-conditioning, ventilation, lightings, and lifts within the company's buildings in Singapore. With the goal of ensuring environmental protection, a broad spectrum of measures was put in place under the airline's Office Management System (OMS) (Singapore Airlines, 2015).

During the 2014/15 financial year, Singapore Airlines buildings and facilities were upgraded with more energy-efficient equipment and technology. These energy efficiency measures included the upgrading of lifts with a more efficient model that had a variable voltage variable frequency (VVVF) motor, the changing of high bay lights from metal halide lamps to more energy-efficient light emitting diode (LED) lights, the replacement of neon signs on the company's main buildings with LED technology, the installation of a heat exchange system at the SilverKris Lounge at Changi Airport, which is used to produce hot water for the showers, and the replacement of an old chiller system with a higher energy efficiency model. These energy saving measures amounted to 80,000 kWh per month (Singapore Airlines, 2015) and this energy saving measure reduces carbon dioxide (CO_2) emissions by around 68.83 tonnes per month. In the 2016/17 financial year, Singapore Airlines energy saving measures primarily focused on the management of air conditioning systems and lighting within its buildings. A key measure was the replacement of high bay lighting for hangar operations (Singapore Airlines, 2017). These measures reduced both energy consumption and the associated carbon dioxide (CO_2) emissions. In the 2017/18 financial year, Singapore Airlines continued its energy saving measures program which included the completion of replacement of high bay lights project for the airline's Hangar 1 from 1,000W sodium metal halide lamps to 400W dimmable LED high bay lights. This measure provided energy efficiency savings and lower carbon dioxide (CO_2) emissions. Other energy efficiency measures included the ongoing replacement of fan coil units (FCUs) within the company's buildings with more efficient models as well as the replacement of computer air-conditioning units (CAUs) serving the aircraft simulator computers with higher efficiency units (Singapore Airlines, 2018b). In the 2018/19 financial year, Singapore Airlines conducted a study that focused on the potential use of solar energy to support the company's use of "green" energy to meet its buildings' energy requirements (Singapore Airlines, 2019). Green energy is "a sustainable energy source that zero or minimal environmental and economic impact, and can be obtained from solar, wind, hydro, geothermal, biomass, and other renewable sources" (Enescu, 2019). A tender was subsequently issued for the installation of solar panels at the airline's Head Office building in Singapore (Singapore Airlines, 2019). Importantly, increasing the proportion of renewable energy consumption to replace part of the fossil energy consumption is regarded as very suitable solution for the problem of air pollution (Shen et al., 2020). This is because renewable energy sources reduce air pollution and cuts down carbon dioxide (CO_2) emissions (Spellman, 2015; Xie et al., 2021). Furthermore, the use of solar power helps a firm to mitigate its greenhouse gases (GHGs) and achieve it sustainability objectives (Sreeneth et al., 2021). Other energy saving measures in the 2018/2019 financial year included the progressive upgrading of air-conditioning equipment to ensure that the most energy efficient equipment is used in the company's buildings and facilities. Singapore Airlines also continued the replacement of Fan Coil Units (FCUs) within its buildings with more efficient models, and the company continued to replace old light fittings with LED lighting (Singapore Airlines, 2019).

In the 2019/20 financial year, Singapore Airlines continued to implement energy reduction initiatives, and these resulted in an estimated 800 MWh of energy savings principally through adjustments to the operating parameters of its chiller plants and air-handling units (AHU), and through the replacement of lighting with more energy efficient light -emitting diode (LED) light fittings. In addition to its energy reduction initiatives, the airline started using renewable energy. As part of this strategy, Singapore Airlines entered a partnership with SembCorp Solar to install rooftop solar panels on three of its buildings: Airline House, SIA Training Centre and TechSQ. As well as delivering considerable energy savings, the new system has reduced the airline's carbon dioxide (CO_2) emissions by 4.3 million tonnes per year (Singapore Airlines, 2021).

In the 2021/2022 financial year, Singapore Airlines maintained its energy efficiency measures program. Consequently, an extensive number of energy efficiency measures were undertaken. These energy efficiency measures included the upgrade and adjustment of the operating parameters of one of its chiller plants and air-handling units (AHU). The airline reduced unnecessary usage of utilities such as air conditioners, lighting, and IT hardware in the SIA Data Centre: The data centre used more energy efficient air-conditioning and computer hardware. In addition, the airline progressively replaced lights with more energy efficient light-emitting diode (LED) light fittings. During the 2021/2022 financial year, all the conventional metal halide high bay lights in the SIA Supplies Centre were retrofitted with energy efficient LED high bay lights. The use of energy efficient LED high bay lights is expected to save 330,000kWh annually, which is equivalent to 135 tonnes of annual carbon dioxide (CO_2) emissions savings. In the 2021/2021 financial year, Singapore Airlines also replaced goods and passenger lifts in the SIA Supplies Centre. Also, in the 2021/2022 financial year, Singapore Airlines let a tender for the replacement of chiller plant systems in the SIA Training Centre and TechSQ facilities. To optimize operating efficiency, existing chiller systems, which were no longer operating at its optimal performance and efficiency, will be combined into a larger and more efficient chiller system. The combined chiller system is expected to save more than 700,000kWh of energy annually. This will result in an annual reduction of 286 tonnes of carbon dioxide (CO_2) emissions. The new chiller system is anticipated to be commissioned in the fourth quarter of 2022/23 financial year (Singapore Airlines, 2022).

4.5.5. Improved Flight Operations Procedures

A key focus for Singapore Airlines environmentally sustainable operations has been the adoption of several improved flight operation measures that not only reduce aircraft fuel consumption but also the associated carbon dioxide (CO_2) emissions. These measures include the use "continuous descent" operations and are designed to minimize fuel use (without impacting safety) (Singapore Airlines, 2017, 2018). A "continuous descent" operation is one in which the arriving aircraft descends continuously, to the greatest possible extent, by employing minimal engine thrust, prior to the final approach fix/final approach point (Young, 2018).

Operational procedures that reduce fuel burn for Singapore Airlines Airbus A380 operations have been implemented at London's Heathrow Airport. Singapore Airlines Airbus A380 aircraft departing from Heathrow Airport use less power when taking off, resulting in fuel savings and less carbon dioxide (CO_2) and nitrous oxide (NOx) emissions. Furthermore, Singapore Airlines optimization of flight operations and flight planning systems have led to more optimum flight route selection (Singapore Airlines, 2017, 2018b), and thus, a reduction in carbon dioxide (CO_2) emissions.

In the 2021/2022 financial year, Singapore Airlines route structure for flights arriving at Singapore Changi Airport were reviewed, and, as a result, direct tracks on applicable flight routes enabled the airline to achieve better route efficiency. This initiative resulted in Singapore Airlines saving around 456 tonnes of fuel and 1,436 tonnes of carbon dioxide (CO_2) emissions annually (Singapore Airlines, 2022).

4.5.6. Improving Aircraft Zero Fuel Weight (ZFW) Used to Plan Aircraft Fuel Uplift

An aircraft's maximum zero fuel weight (MZFW) is the maximum weight permitted on the aircraft before fuel is loaded. The MZFW is limited by the strength and aircraft airworthiness requirements (Jofré & Irrgang, 2000). Importantly, the maximum zero fuel weight (MZFW) defines the available payload capability on a given flight (Meijer, 2021). By improving the accuracy of an aircraft's maximum zero fuel weight (MZFW), the correct amount of fuel is uplifted on the flight. This reduces the overall weight of the aircraft. Singapore Airlines has implemented a range of measures to improve the MZFW accuracy of its aircraft fleet. The airline has deployed a "ZFW Monitoring Dashboard", which enables its Airport Operations department to monitor the MZFW of its aircraft fleet more closely. In the 2019/20 financial year, the optimization of aircraft MZFW resulted in an estimated 1,800 tonnes of fuel savings, which translated to an estimated annual saving of 5,670 tonnes of carbon dioxide (CO_2) emissions (Singapore Airlines, 2020).

4.5.7. Modification to the Airbus A380 Trent 900 Engines

To enhance the performance of its fleet of Airbus A380 aircraft, Singapore Airlines has modified the aircraft's Rolls-Royce Trent 900 engines, which has resulted in annual savings of 10,010 tonnes of fuel and an annual reduction of 31,531.5 tonnes of carbon dioxide (CO_2) emissions (Singapore Airlines, 2017, 2018b).

4.5.8. Removal of Economy Class Footrests in Selected Aircraft

In 2019, Singapore Airlines removed some Economy Class footrests on selected aircraft. This initiative saved around 200kg to 300kg per aircraft (Singapore Airlines, 2019). The lower aircraft weight translates into lower fuel burn, and thus, lower emission levels.

4.5.9. Singapore Airlines' "Green Package" Flights

In May 2017, in accordance with the airline's commitment to reduce international aviation emissions, Singapore Airlines partnered with the Civil Aviation Authority of Singapore (CAAS) and operated a series of 12 "green package" flights over a three-month period on the airline's non-stop San Francisco-Singapore route. These flights utilized Singapore Airlines latest-generation and most fuel-efficient aircraft – the Airbus A350-900XWB aircraft – as well as sustainable aviation fuels, and optimized air traffic management best practices (Singapore Airlines, 2019).

The 12 "green package" flights were powered by a combination of hydro-processed esters and fatty acids (HEFA), a sustainable biofuel produced from used cooking oils and conventional jet fuel. This fuel offers a life cycle potential reduction of 80% in carbon emissions when compared to traditional jet fuel (Singapore Airlines, 2019).

4.5.10. The Optimization of Aircraft Fuel Efficiency

An important characteristic of the global airline industry is that it is highly energy intensive (Baxter et al., 2021) and fuel intensive. As a result, high and volatile aircraft fuel prices can have a significant impact on an airline's bottom lines (Liu, 2021). Nowadays, airlines are placing a high focus on jet fuel cost and aircraft fuel efficiency. The fuel cost is typically the largest cost for most airlines (Zhang & Zhang, 2018). Reducing aircraft fuel consumption affects an airline's bottom line by reducing fuel costs and the related carbon dioxide (CO_2) emissions (Johnson & Gonzalez, 2013). Thus, aircraft fuel efficiency has become one of the most important policy goals for airline operations management (Oliveira et al., 2021). Indeed, growing environmental concerns have attracted significant attention of the air transport industry in recent times towards the requirement for the judicious use of aviation fuel (Singh et al., 2018). Airlines and the aircraft manufacturers have invested in new technologies and strategies to reduce aircraft fuel consumption, and thus, reduce their carbon dioxide (CO_2) aircraft emissions. Aircraft fuel has a close relationship with the emissions of carbon dioxide (CO_2) (Zou et al., 2016).

Singapore Airlines has placed a high focus on its aircraft fuel efficiency and has implemented a range of measures that are designed to optimize its aircraft fuel efficiency. The added benefit of such a strategy is the reduction in carbon dioxide (CO_2) emissions due to the lower amounts of fuel used on both its passenger and air cargo services. In the 2021/2022 financial year, Singapore Airlines carefully monitored its fuel usage to ensure an optimal level of uplifted fuel on its aircraft. The airline also sought to optimize its contingency fuel to reduce fuel uplift. Singapore Airlines also utilized more fuel-efficient aircraft and engines for its long-haul. This practice reduced the amount of fuel burnt during a flight. Also, in the 2021/2022 financial year, Singapore Airlines ensured that optimal flaps settings were used during take-off to reduce the amount of fuel burnt (Singapore Airlines, 2022).

4.5.11. The Use of Aviation Biofuels

Prior to examining Singapore Airlines sustainable fuel strategy, it is important to note that the concerns related to climate change and energy supply have resulted in the production of more sustainable aviation fuels (SAF) (Brooks et al., 2016). As a result, there has been a growing interest by the global aviation industry in sustainable aviation fuels (SAF) (Trejo-Pech et al., 2019). Accordingly, sustainable aviation fuels are increasingly being viewed as an ideal option for the airline industry to achieve large, near-term emissions reductions (Staples et al., 2014). This is because the use of biomass-and crop-based sustainable aviation fuels can help reduce emissions in the aviation industry (McCollum et al., 2021). Consequently, airlines are now using aviation biofuel as an environment sustainability measure (Baxter et al., 2020; Goh et al., 2022). Depending upon the raw material used in its production, biofuels can reduce carbon dioxide (CO_2) emissions by 60-80% (Bioenergy International, 2019; Tavares Kennedy, 2019).

Singapore Airlines has been an active member of the "Sustainable Aviation Fuel Users Group" (SAFUG) since 2011. SAFUG was established to accelerate the development and commercialization of Sustainable Aviation Fuels (SAF). Singapore Airlines views sustainable aviation fuels as a key long-term measure to support the air transport industry's carbon-neutral growth goal beyond 2020. Accordingly, the airline has pledged to advance and adopt aviation biofuels produced in a sustainable way. The sustainable aviation fuels should have minimal impact on biodiversity, such fuels should meet a sustainability standard, in relation to land, water and energy use, the production of sustainable aviation fuels should not displace or compete with food crops; and these fuels should also provide a positive socio-economic impact (Singapore Airlines, 2019).

In addition to the use of sustainable aviation fuels on its series of "Green Flights" in 2017, Singapore Airlines began working with Stockholm's Swedavia Airport in 2020 on the use of sustainable aviation fuels (Becken, 2021). In January 2020, Singapore Airlines began a year-long partnership with Swedish airport operator Swedavia. As part of Swedavia's SAF Incentive Scheme, the airline commenced using a blend of jet fuel and SAF on its flights between Stockholm and Moscow (Singapore Airlines, 2020). This arrangement between the two parties continued in the 2020/21 financial year and resulted in a reduction of around 47 tonnes of carbon dioxide (CO_2) (Singapore Airlines, 2021).

In the 2021/2022 financial year, Singapore Airlines together with Civil Aviation Authority of Singapore (CAAS) and Temasek conducted a year-long pilot study that focused on ways to operationalize the deployment of blended sustainable aviation fuel (SAF) at Singapore Changi Airport (Singapore Airlines, 2022).

In February 2022, Singapore Airlines firmed up orders for 1.25 million litres of neat sustainable aviation fuel (SAF), which will be blended with refined jet fuel and delivered to Singapore Changi Airport for use on Singapore Airline and its subsidiary Scoot Tigerair flights from the third quarter of 2022. This sustainable aviation fuel (SAF) pilot program is anticipated to reduce the two airlines carbon dioxide (CO_2) emissions by around 2,500 tonnes per annum (Singapore Airlines, 2022).

In February 2022, Singapore Airlines was the first airline to sign the Global Sustainable Aviation Fuel (SAF) Declaration (Polek, 2022; Singapore Airlines, 2022; Tolba, 2022). The Global SAF Declaration calls on industry partners from the aerospace, aviation, and fuel value chains to jointly work together to increase the uptake of sustainable aviation fuel (SAF) as an important part of the industry's decarbonization with the ambition of ensuring a steady ramp up in the use of sustainable aviation fuel (SAF) over the next decade (Travel News Asia, 2022). As previously noted, sustainable aviation fuel (SAF) offers a considerably smaller carbon dioxide (CO_2) emission footprint when compared to the use of fossil fuels (Shahriar & Khanal, 2022).

4.5.12. The Use of Light Weight Flight Catering Items on Regional Services

In 2020, Singapore Airlines introduced a new regional economy class which offered passengers increased meal choices and these were accompanied with bamboo cutlery and sustainable paper packaging. The new packaging was lighter, weighing half that of plastics. This reduction in weight resulted in lower fuel consumption (Becken, 2021; Singapore Airlines, 2021), and thus, lower carbon dioxide (CO_2) emissions.

4.5.13. The Use of Lightweight Air Cargo Containers

An important development in the global airline industry in recent times has been the use by airlines of light weight aircraft containers or unit load devices. These containers help to minimize aircraft weight without compromising the business volume of the lightweight aircraft unit load devices (ULDs) (Laniel et al., 2011). Aircraft unit load devices, or ULDs, are pallets and containers which are used to carry air cargo, mail and passenger baggage on wide-body passenger and freighter aircraft (Baxter et al., 2014; Lu & Chen, 2011). The trend towards increased use of lightweight ULDs reflects airline's objective of achieving fuel savings and reducing the carbon dioxide (CO_2) impact within the industry (Bandi & Lumia, 2013). Singapore Airlines is one such airline that has acquired light weight aircraft ULDs that contribute to its ability to reduce carbon dioxide (CO_2) emissions (Singapore Airlines, 2021).

4.5.14. The Use of Mobile Ground Power Units

Electrical power is required on the airport apron for the servicing of aircraft prior to engine start-up. (Ashford et al., 2013; Horonjeff et al., 2010; Kazda & Caves, 2015). As part of their environmental policy, Singapore Airlines uses mobile ground power units and preconditioned air units during night layovers and long transits to reduce the reliance on its aircraft auxiliary power units (APUs) (Singapore Airlines, 2017, 2019). This reduction in fuel consumption also translates into lower carbon dioxide (CO_2) emissions. In the 2019/20 financial year, this measure delivered an estimated 910 tonnes in fuel savings, which translated into an annual saving of 2,867 tonnes of carbon dioxide (CO_2) emissions (Singapore Airlines, 2020).

4.5.15. Total Mission Management (TMM)

In the 2020/21 financial year, Singapore Airlines Total Mission Management (TMM) Department collaborated with the airline's Flight Operations Division and the Civil Aviation Authority of Singapore (CAAS) to introduce various initiatives whose objective was to improve operational efficiencies (Singapore Airlines, 2021). Singapore Airlines Total Mission Management (TMM) is a transversal department that works very closely with cabin crew, pilots, engineers, and airport staff to improve the airline's operational excellence and flight punctuality (Lim, 2020). As part of this collaboration, the workgroup also explored opportunities to implement "green" operating procedures using a data driven approach. This objective was achieved through the development of data analytics tools from Rolls-Royce for insights on flight data, together with regular meetings with the CAAS to assess Air Traffic Management (ATM) performance and to identify initiatives to improve flight efficiency of the Singapore air hub. As part of this collaboration, a range of 'green' operational initiatives was implemented, which resulted in the improved efficiency of flight operations in the Singapore airspace (Singapore Airlines, 2021).

At times, Singaporean airspace is often congested. As a result, flights may be required to hold for longer periods of time before landing, this leads to extra fuel burnt. To reduce the aircraft arrival holding periods, the TMM Department worked with the CAAS and the Singapore Airlines Flight Operations Division to sequence flights before they enter Singapore airspace to better provide adequate separation between flight arrivals (Singapore Airlines, 2022). This initiative resulted in an annual reduction of 1,808 tonnes of carbon dioxide (CO_2) emissions (Singapore Airlines, 2021).

At the time of the present study, the TMM Department was still collaborating with the CAAS and the airline's Flight Operations Division to review the route structure for flights arriving at Singapore. Direct flight tracks on applicable flight routes were introduced to achieve better aircraft route efficiency. In the 2020/21 financial year, an initial trial was carried out between September 2020 and January 2021 to establish the process, and this trial was subsequently extended to November 2021. The trial was subsequently extended to include new routes. This initiative reduced Singapore Airlines annual carbon dioxide (CO_2) emissions by 599 tonnes (Singapore Airlines, 2021).

Due to the normally high air traffic density in Singapore's airspace, flights arriving in Singapore are typically subjected to step descents. This practice results in higher aircraft fuel burn. During the 2020/21 financial year, the TMM Department worked closely with the CAAS to establish a procedure that reduces the steep descent for flights entering the Singapore airspace, and this in turn reduces aircraft fuel burn. This initiative had favorable an environmental impact as it reduced Singapore Airlines annual carbon dioxide (CO_2) emissions by an estimated 725 tonnes in the 2020/ 2021 financial year (Singapore Airlines, 2021). A similar situation occurred in the 2021/2022 financial year, when the airline reduced its carbon dioxide (CO_2) emissions by 2,041 tonnes by using this enhanced airspace management procedure (Singapore Airlines, 2022).

The Cost Index (CI) that is used by Singapore Airlines is a ratio that defines the time-related cost versus cost of fuel when operating an aircraft on a flight. This figure is calculated at the pre-flight stage so that the economical speed for the flight can be determined. As part of Singapore Airlines Cost Index Adjustment process, the TMM Department monitors each aircraft's flight time and adjusts the CI at the flight planning stage to achieve on-time-performance whilst also conserving fuel by utilizing a lower CI when flights are estimated to arrive at Singapore ahead of schedule. This initiative reduced Singapore Airlines annual carbon dioxide (CO_2) emissions by around 1,827 tonnes in the 2020/21 financial year (Singapore Airlines, 2021). In the 2021/2022 financial year, Singapore Airlines achieved a saving of 602 tonnes of fuel that was equivalent to 1,896 tonnes of carbon dioxide (CO_2) emissions saved from using this operational efficiency measure (Singapore Airlines, 2022).

4.5.16. Washing of Aircraft Engines

Singapore Airlines has implemented an aircraft fleet-wide engine washing program (Singapore Airlines, 2017). This program has resulted in lower annual carbon dioxide (CO2) emissions of around 32,670 tonnes of carbon dioxide (CO_2). Since the 2018/19 financial year, this initiative has been implemented by the airline for their Rolls-Royce Trent XWB and Trent 1000 engines on the airline's fleet of Airbus A350-900XWB and Boeing 787-10 aircraft respectively. As of 31 March 2022, 148 tonnes of fuel, equivalent to 466 tonnes of carbon dioxide (CO_2) emissions, had been saved from optimized engine washing in the 2021/22 financial year (Singapore Airlines, 2022).

4.6. Singapore Airlines Passenger Carbon Offsets Program

Singapore Airlines (SIA) and its low-cost carrier (LCC) Scoot subsidiary introduced a voluntary carbon offsets program in June 2021. This program enables the airline's passengers to offset their share of the flight's carbon emissions by contributing to environmental projects in Indonesia, India, and Nepal. Under the program, passengers can purchase carbon offsets from dedicated microsites before or after a flight. From late 2021, the airline's passengers would be able to use their "KrisFlyer" miles and "HighFlyer" points to purchase carbon offsets (Surgenor, 2021; The Business Times, 2021). The airline's carbon offset environmental projects include the protection of forests in Indonesia, supporting renewable solar energy projects in India, and the provision of efficient, clean burning cookstoves for rural families in Nepal (Baker, 2021; Otley, 2021; Surgenor, 2021).

V. CONCLUSION

In conclusion, this study has investigated Singapore Airlines annual carbon footprint as well as the strategies defined and implemented by Singapore Airlines to mitigate the environmental impact from its flights, ground operations, and Singapore-based facility and building energy consumption related carbon dioxide (CO_2) emissions. To achieve the objectives of the study, Singapore Airlines was selected as the case airline. The research was underpinned by an in-depth qualitative longitudinal case study research approach. The study's qualitative data was examined using document analysis. The study was supported by a case study research framework that followed the recommendations of Yin (2018).

Singapore Airlines has defined and implemented a very comprehensive environmental policy and has pledged to achieve net zero carbon emissions by 2050. This goal will be attained through the reinforcement of its long-held strategy of working towards decarbonization and environmental sustainability across its global operations. The case study also revealed that Singapore Airlines is an active participant in the "Singapore Green Plan 2030", and thus, the airline is aligning its green objectives with the "Singapore Green Plan". Singapore Airlines has also embraced the International Air Transport Association (IATA) climate targets, and this is underpinned by the four pillars of Singapore Airlines environmental policy.

The case study found that Singapore Airline's carbon footprint is comprised of its Scope 1 emissions that are produced from the provision of passenger and air cargo services together with the carbon dioxide (CO_2) emission produced from its ground operations, and the Scope 2 indirect emissions which are produced from the airline's consumption of electricity at its Singapore hub. Singapore Airlines principal sources of carbon dioxide (CO_2) emissions is from its passengers' services and its consumption of electricity. A key finding of the case study is that despite Singapore Airlines growing the number of passengers carried, the number of destinations served and its aircraft fleet size, the annual Scope 1 carbon dioxide (CO_2) emissions have developed at a rate that is much lower than the growth in enplaned passengers. This is a very favorable trend that was helped by the extensive carbon dioxide (CO_2) reduction measures implemented by the airline over the study period. Over the study period, Singapore Airlines Scope 2 carbon dioxide (CO_2) emissions exhibited an overall downward trend. This very favorable decrease was positively impacted by the large range of energy efficiency measures implemented by Singapore Airlines throughout the study period. The case study revealed that Singapore Airlines annual Scope 1 carbon dioxide (CO_2) emissions from fossil fuel consumption oscillated throughout the study, reflecting differing usage requirements. In addition, air cargo is a core business product of Singapore Airlines and throughout the study period its cargo division, SIA Cargo, operated a fleet of Boeing 747-400 freighters that linked key air cargo markets with its Changi Airport Hub. The total annual Scope 1 carbon dioxide (CO_2) emissions from freighter services largely exhibited a general downward trend over the study period. This was a very favorable response given the number of services provided by SIA Cargo throughout the study period.

A key finding of the case study is that Singapore Airlines has implemented a wide range of carbon dioxide (CO_2) emissions reduction measures that have focused on the reduction in aircraft weight which delivers fuel burn savings, and thus, lower carbon dioxide (CO_2) emissions, improved operational procedures, optimization of air space management in collaboration with key air traffic control agencies, the use of cleaner energy vehicles, an extensive range of energy efficiency measures in its buildings and facilities, the use of sustainable aviation fuels, and the use of more energy efficient ground power sources when its aircraft have night layovers and long transits. Finally, Singapore Airlines has implemented a passenger carbon offsetting program, which enables passengers to offset carbon dioxide (CO_2) emissions by contributing to projects in Central Kalimantan in Indonesia, solar power projects in India, and a final project that is providing more efficient, clean burning stoves for rural homes located in Nepal.

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Moth Repellent Yellow-Specific Bulbs for the Management of Lepidopteran Pod Borers and Leaf Eating Caterpillars on Vegetable Pest Management Programs in Sri Lanka

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***Abstract*—*The study was conducted research field at Horticultural Crops Research and Development Institute (HORDI), Gannoruwa, Sri Lanka and different farmer fields to determine the efficacy of yellow bulbs during nights for the management of lepidopteran Pod borers and leaf eating caterpillars on brinjal, okra and cabbage. Three research field experiments at HORDI, Gannoruwa during, 2018 Minor (Yala), 2018/2019 Major (Maha) and 2019 Yala and-farmer field demonstrations were conducted to determine the effectiveness of the yellow bulbs for the management of Brinjal Shoot and Pod Borer, Okra pod borer and Cabbage leaf eating caterpillars. Research fields measuring 100 m^2 – 200 m^2 were selected for each treatment at ~~the~~ HORDI research fields and 1,000 m^2 fields for farmer field demonstrations. Yellow LED bulbs (at the rate of nine 1.5W bulbs /1,000 m^2 were installed . Replication were done by dividing each plot into four sub-plots. Lands were prepared and bulbs were installed in the yellow bulb treatments and planting was done each plot. Data were recorded from randomly selected twenty plants from each sub plot for pest damage in weekly intervals. Yield was recorded at harvest to measure the yield loss due to pest damage. Data revealed that fields illuminated with yellow bulbs has given significantly low number of pest damage in each crops. It was reported from the cabbage experiment conducted during yala 2018, significantly low number of caterpillar damage in yellow bulb treatment compared to untreated control and chemical treatment. Similar results gave from the experiment conducted during Maha 2018/2019 for the same crops. Results of the experiment conducted against the okra pod borer during Yala 2019, revealed that significantly low-percentage of pod damage on plots exposed to yellow light. Field trials conducted on farmer fields and research farm further confirmed the results with low number of pest damage on okra and brinjal crops.*

***Keywords*— Specific Yellow Bulb, Okra pod borer, Cabbage caterpillars, Brinjal shoot and pod borer**

I. INTRODUCTION

Damage caused to vegetables by noktunoid lepidopteron larva is a serious issue in sustainable vegetable production in Sri Lanka. Among them okra, tomato and brinjal pod borers and leaf eating caterpillars of Cabbage are the major pests damaging to the marketable harvest of these crops **(Nishantha *et al.*, 2017)**. There are several species of lepidopteron pod borers, such as *Earias vittella*, (the Okra shoot and pod borer), *Helicoverpa armigera*, (the tomato fruit borer) and *Leucinodes orbonalis*, (the brinjal pod borer) . Among the leaf eating caterpillars *Spodoptera litura*, *Hellula undalis*, *Chrysodeixis eriosoma*, *Crocidolomia binotalis* and *Plutella xylostella* are most harmful ~~to~~ pests of cabbage. Farmers indiscriminately use heavy doses of insecticides, especially IGRs for the management of these pests **(Nishantha *et al.*, 2017)**. Therefore, most of theese vegetables available in the market may be contaminated with pesticide residues **(Gapud and Canapi 1994; Orden *et al*., 1994)**.

The use of "yellow-specific lamp to repel caterpillars that are harmful to leaves and fruit/pods" are one of mature technologies use in green houses in Japan to avoid damage caused by these nocturnoid lepidopteran moths. The Most specific features of this yellow color bulbs are Nocturnoid lepidopteron specificity with emitting yellow color, regulated with 570 – 590 nm wavelength, which is detect by the moth during nights and when its blinking during nights, the moth compound eyes determine it as day time and it will stop come into crop for egg laying **(Shimoda M & Honda K, 2013)**.

Therefore, with worldwide information and the published fact on this product, The Horticultural Crops Research and Development Institute of the Department of Agriculture initiated research programmes to introduce "Novel High tech introductions for pesticides free Agriculture" for the country in management of pests specially pod borers and leaf eating caterpillars without using-pesticides.

The experiments were started on research fields at HORDI and a few farmer fields to test the effectiveness of the yellow light for managing **nocturnoid** Lepidopteran pest of Cabbage, brinjal and okra during growing seasons of 2018 *Yala*, 2018/2019 *Maha* and 2019 *Yala*.

II. METHODOLOGY

2.1 Research Field Experiments

Three seasons experiments were conducted for cabbage and okra. Three large plots were prepared, each with 100 m^2 – 200 m^2 area. Each plot was divided into four sub plot for replication. Yellow bulbs were installed as the First treatment at the rate of 9 bulbs (1.5 W) /1,000 m^2 (570-590 nm/ emitting color yellow- blinking) and insecticide treated and untreated control plots were also maintained.

During the bulb installation, the distance between two bulbs were kept 7 to 10 meters and height of the bulbs from the crop canopy was kept about 2 to 2.5 m above from the crop (Plate 01 and Plate 02). All the plots treated with yellow bulbs were established with 40 mesh insect proof nets around the field to avoid entering of other pest species damaging to these crops. Crops were established and lights were switch on every day at 6.00 pm and switch off on the following day at 6.00 am. The plots designated for insecticide applications were treated with recommended insecticides in seven day intervals.

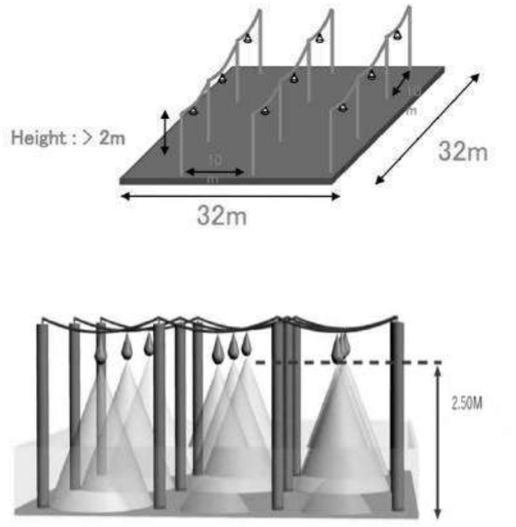

Plate 01: Dramatic Installation of the installation technique of the yellow bulbs for 1000 m^2 area, field layout and height of the bulbs to installed

2.2 Farmer Field Experiments

Three experiments were conducted in three locations in three different time periods. Three large plots were prepared, each measuring 1,000 m^2. Nine bulbs were installed according the layout given in the plate 01 and plate 02. Physical barriers for other pests were made by establishing forty mesh insect proof nets were placed around the field and crops were established. Lights were switch on every day at 6.00 pm and switch off following day 6.00 am. Insecticide treatment were treated with recommended insecticides in seven day intervals. Data were recorded all the treatments according the procedure followed at research field experiments. All treatments were replicated three times, and the crop yields, insect populations were subjected for ANOVA.

III. RESULTS

3.1 EXPERIMENT 1: Effect of yellow-specific bulbs to repel Cabbage caterpillar complex during *Yala* 2018 season

The data on Table 01 revealed that highest number of larva were recorded from untreated control and it was significantly higher than the other treatments. The lowest number of larva were recorded from yellow bulb and it was not significantly different to the chemical control. Larger population of *Crocidolomia binotalis* and second highest was *S. litura*. However, the most serious pest, *P. xylostella*, diamondback moth caterpillar larva was recorded in lowest numbers this season in all the treatmented plots

According to results (table 02) highest yield was recorded from the plots exposed to Yellow bulb treatment (39.375 t/ha) and lowest was recorded from the Chemical control treatment (14.676 t/ha). Marketable yield (28.035 t/ha), marketable leaf weight (8.325 t/ha) were highest on treatments with yellow bulb and lowest was from plots treated with Flubendiamide 20% SG. The main reason for lowest yield from chemical treatment field was due to prevailed water logging condition of the plot. Therefore, plants were not grown well and heads were not formed properly.

Table 01 - Total Number of different types of larvae per ten on cabbage crop on different treatments - Yala 2018 season HORDI, Gannoruwa

Treatment	***Spodoptera litiura****	***Crocidolomia binotails****	***Crisodeixis erisoma****	***Plutella xylostella****
T_1 - Yellow bulb	2.5	2.25	1.25	1
	(1.6975 b)	(1.6075 b)	(1.1900 a)	(1.0625 b)
T_2 - Flubendiamide 20% SG (recommended rate)	4.0	4.25	1.5	0
	(2.10 b)	(1.9425 b)	(1.2475 a)	(0.7100 b)
T_3 - untreated Control	10.0	70.0	1.5	10.5
	(3.1925 a)	(8.335 a)	(1.2475 a)	(3.2850 a)
CV	18.26	27.69	39.75	33.84

*$\sqrt{X+0.5}$ values were subjected to analysis and are given within the brackets

* Mean values within a column followed by the same letter(s) are not significantly different at $p<0.05$ based on the LSD test

Table 02 - Yield data of the cabbage crop at the end of the Yala 2018 season (HORDI, Gannoruwa)

Treatment	**Total weight (t/ha**	**Marketable yield (t/Ha)**	**Marketable leaf weight (t/Ha)**	**Undamaged weight (t/Ha)**	**Damage weight (t/Ha)**
Yellow bulb	39.375	28.035	8.325	36.362	3.013
Flubendiamide 20% SG	14.676	6.585	5.089	11.674	3.001
Control	25.335	12.165	6.428	18.594	6.741

Table 03 - Summary of the different types of larvae per ten plants in each treatment during Maha 2018/2019 (HORDI, Gannoruwa)

Treatments	***Spodoptera litura***	***Crocidolomia binotails***	***Chrysodeixis erisoma***	***Plutella xylostella***	***Aphids***
Yellow bulb	4.00	1.00	0.33	-	235.67
	(2.11 a)	(1.17 a)	(0.88 a)		(15.25 b)
Azidaractin 5% EC	12.67	1.67	2.67	-	62.67
	(3.61 bc)	(1.26 a)	(1.77 bc)		(7.81 a)
Spinosad 2.5 % SC	14.33	6.33	2.00	-	94.67
	(3.85 c)	(2.61 c)	(1.56 b)		(9.73 a)
Flubendiamide 20 % SG	6.00	5.67	3.67	-	111.00
	(2.51 ab)	(2.48 bc)	(2.04 c)		(10.43 a)

Control untreated	18.33 (4.22 c)	59.00 (7.67 c)	10.00 (3.24 d)	-	630.33 (24.97 c)
CV	19.44	23.30	11.15	-	15.88

*√X+0.5 values were subjected to analysis and are given within the brackets

* Mean values within a column followed by the same letter(s) are not significantly different at $p<0.05$ based on the LSD test

The data on Table 03 indicating highest number of larva from untreated control and it was significantly higher than the other treatments. The lowest number of larva were recorded from yellow bulb and it was significantly lower than the chemical control and untreated control. Highest number of caterpillar type was *C binotalis* and second highest was *S litura*. Most importantly no diamondback moth caterpillar larva was recorded from any treatment in this season too. However, in addition to the larval caterpillars, aphids were recorded in higher numbers in all the treatments. This was significant in yellow bulbs treatment (235.67) compared to other insecticides treatments and significantly lower compared to the untreated control (630.33).

Table 04 - Summary of the number of different types of natural enemies per ten plants in each treatment during Maha 2018/2019 HORDI Gannoruwa

Treatments	***Natural enemies of aphids***	***Spider spp.***	***Ladybird spp.***
Yellow bulb	20.67 (3.97 a)	4.67 (2.11 a)	0.66 (1.05 a)
Azidaractin 5% EC	5.67 (1.87 a)	2.67 (1.61 a)	0.00 (0.71 a)
Spinosad	2.00 (1.48 a)	4.67 (2.27 a)	0.33 (0.88 a)
Flubendiamide 20% SG	29.33 (3.94 a)	6.33 (2.61 a)	1.33 (1.29 ab)
Control	12.67 (3.53 a)	5.33 (2.39 a)	3.33 (1.89 b)
CV	99.00	32.52	34.44

*√X+0.5 values were subjected to analysis and are given within the brackets

* Mean values within a column followed by the same letter(s) are not significantly different at $p<0.05$ based on the LSD test

According to data on table 04, higher number of natural enemies were recorded from yellow bulb treated fields and it was not significantly different from other four treatments. However, highest number natural enemies as aphids' natural enemies. It may be due to high population of aphids record on yellow bulb treatment. Natural enemies recorded can be categorized as surphid flies, ladybirds, spiders and parasitoid, *aphidius* spp. Highest yield loss was recorded from untreated control treatment (Fig 01) and lowest yield was recorded from yellow bulb treatment. However, yield loss due to chemical treatments were also reduced compare to the untreated control and it was comparatively high compare? to the yellow bulb treated plots.

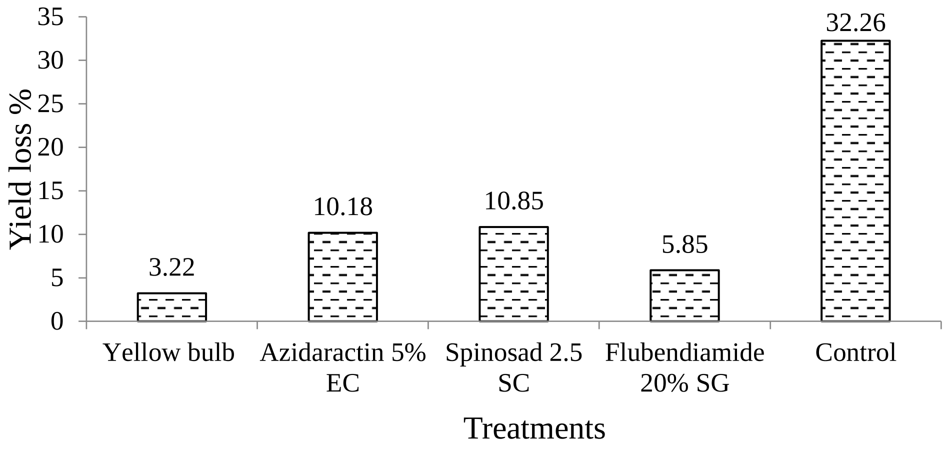

Fig.1: Percentage yield loses at the end of the Maha 2018/2019 season on cabbage

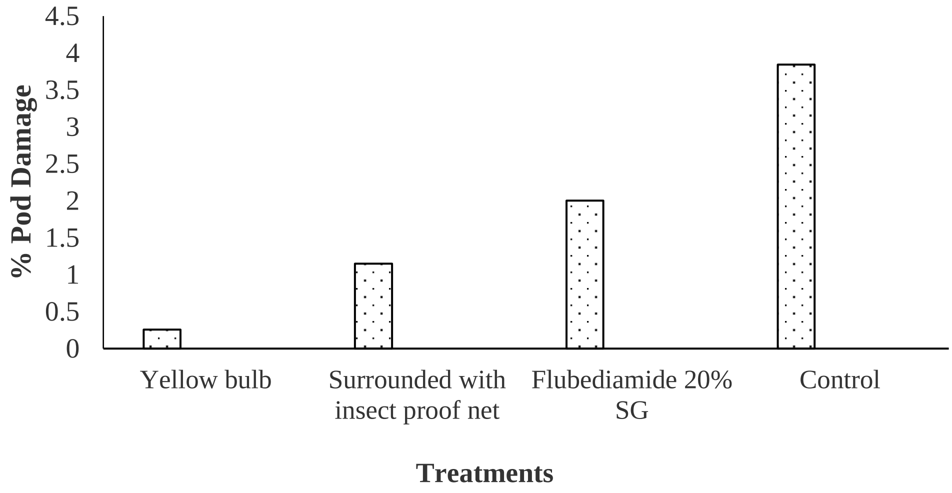

Fig.2 - Percentage Pod damage at the end of the Yala 2019 season in each treatment on okra crop

3.3 EXPERIMENT 3: Effect of Use of yellow-specific bulbs to repel okra pod borer, *Earias vitella* during Yala 2019

The okra trial in Yala 2019 reveled the number of pod damage (Figure 02) plots treated with yellow bulbs gave lower yield loss compared to the other treatments. However, highest pod damaged was recorded from untreated control and it was significantly higher compared to Yellow bulbs and chemical control treatment.

3.4 FARMER FIELD DEMONSTRATIONS

Table 05 - Effect of yellow-specific bulbs to repel different caterpillars on different seasons at farmer fields

Location	Season/time period	Treatment	Total yield (Kg/1000m^2)	Damage yield (Kg/1000m^2)	% Damage
Manik~~k~~hinna, Kandy	*Yala* 2022	Yellow bulb	719	48	6.67
		Control	373	45.6	12.23
Palugaswawa, Anuradhapura	*Yala* 2022	Yellow bulb	346.5	3.5	1.01
		Control	257.2	31.3	12.17
Madurankuliya,	*Yala* 2020	Yellow bulb	691.6	18	2.60

Kalpitiya		Control	691.25	51.38	7.43
Gampaha, Meerigama	*Yala* 2018	Yellow bulb	2975	0	0
		Control	1785.2	175.8	9.84

Farmer field demonstrations further confirm the results of the research field experiments, that plots treated with yellow bulbs has given significant reduction in yield losses. Data from all the five locations indicate that higher yield from yellow bulb treatment compared to the control treatment.

Plate 02: Cabbage Field during Yala 2018

Plate 03: Cabbage Field during Maha 2018/2019

Plate 04: Okra Field during Yala 2019

Plate 05: Brinjal Field during Yala 2020

Plate 06: Field layout of the farmer field Meerigama

IV. DISCUSSION

According to **Nomura (1967)**; **Nomura *et al*. (1965)**, Fruit-piercing moths such as *Eudocima tyrannus* Guene and *Oraesia emarginata* Fabricius damage fruit in orchards in Japan.

They were able to manage the damage by successively using yellow fluorescent lamps in the orchard at night. Therefore, this strategy was clearly explained by **Meyer-Rochow, 1974; Walcott, 1969** that when moths encounter light above a certain brightness at night, under which their compound eyes are light-adapted as in the daytime. The light adaptation suppresses nocturnal behaviors such as flying and mating.

Therefore, in this study effect of yellow LED bulbs for the management of Pod and fruit borers and leaf eating caterpillars has proven the success. In this study we were manage major pest of okra, brinjal and cabbages under yellow bulbs. According to **Yase *et al.* 1997** and **Yase *et al.* 2004** this technique has also practice to prevent damage to chrysanthemums and carnations by the cotton boll worm *H. armigera*, damage to green perilla by the common cutworm *S. litura* , and damage to cabbage by the webworm, *H. undalis* .

It is also elaborate from the references from **Yamada *et al*. 2006**; **Kono and Yase 1996**; **Yase *et al*. 1997,** recently, green fluorescent lamps have also been developed for the control of nocturnal moths. Agreeing to them, these lamps suppress the behavioral activity of a number of moth species in the same way as yellow fluorescent lamps but have little effect on the growth of plants compared to the yellow lamps. Consequently, LED lighting is becoming considerably cheaper and therefore, it is becoming very practical and cheaper method for the yellow-emitting LEDs applied to control the behaviors of nocturnal moths (**Hirama *et al*., 2007; Yabu 1999; Yoon *et al*. 2012**). Also with the possibility of LEDs can produce highly unicolor lights (i.e., with a narrow range of wavelength) through the spectrum from UV to red, this character of LEDs is an advantage for controlling pest behavior and their practical application is expected in the near future **(Shimoda & Honda, 2013)**.

V. CONCLUSIONS

The field experiments conducted in three seasons, indicated that the moth repellent yellow bulbs successfully control the Lepidopteran Pod Borers and leaf eating caterpillars. These bulbs found to be effective if they are illuminated throughout the cropping season according to the given recommendations. Field observations has shown no side effects on the other organisms especially natural enemies in the environment. Therefore, the technology can be introduced as a component of Integrated Pest Management programs, it also can be recommended to use under organic farming system and ecofriendly agriculture systems as a component of pest management tool.

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Evaluating the effect of Varying Drying Air Temperatures on Quality Attributes of Avocado (*Persea Americana*) Peels

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***Abstract*— Avocado (Persea americana) belongs to the family of Lauraceae. The processing of this fruit, as well as its fresh consumption, results in large amounts of waste, such as peels and seeds. The peels were dried at 40, 50, 60, 65 and 70 °C using oven dryer with a constant air velocity 1.4 m/s and open sun drying. The bioactive (flavonoids, total phenols and vitamin C) and nutritional content (carbohydrate, fat, fibre, protein, energy and ash content) of the avocado peel was investigated. The drying process took place mainly in the falling rate period indicating that water removal at the initial stage of the drying process was high and there was a rapid decrease as drying continued until equilibrium was reached. The ash, fibre, protein, vitamin c, flavonoids and metabolic energy content increase rapidly with an increase in the drying temperature. There was decrease in the fat, total phenolic content and moisture content of the dried avocado peels in this research. The Nutritional results obtained showed avocado peels had high contents of carbohydrate (40.732 % at 70 °C), fibre (35.187 % at 60 °C) and phenols (25.643 % at 50 °C) and therefore could be used as alternative sources of nutrients and also be added in foods avoiding waste and adding value to the fruit. The results showed that the peels are rich in antioxidants, fibre, flavonoids which helps to fight free radical damage and cancer in mammals and could be use as additive in foods.*

***Keywords*— Avocado, Lauraceae, antioxidants, fibre, flavonoids.**

I. INTRODUCTION

Avocado (*Persea americana*) tree produces a large, fleshy, pear-shaped fruit with a single large oblate seed with two cotyledons enclosed in a thin, brown, papery seed coat often adhering to the flesh cavity; itcomes from the family of *Lauraceae*, genus *Perseal*, subgenus *Persea* and *Eurodaphne* (Duarte *et al*., 2016; Morton, 2020). The avocado fruit is a drupe constituted by epicarp (peel), mesocarp (pulp), and endocarp (seed). Research has proven that the avocado fruit contains essential micro-nutrients for human consumption such as vitamins, minerals and polyphenols. The oil of the fruit has medicinal properties and the peels contains significant amounts of minerals (Rotta *et al*., 2016). The fruit is considered a major topical fruit because it contains omega fatty acid and fat-soluble vitamins D and E at median levels and vitamins A and B which are lacking in other fruits (Duarte *et al*., 2016). The avocado fruit is a perennial plant that can be grown on rough locations and is highly perishable; in addition to its natural perishability, several factors such as mechanical damage, compression and cut, physiological, chemical and biochemical changes are responsible to changes in colour, aroma, taste and texture of the fruit (Duarte *et al*., 2016).

Duarte *et al*. (2016) reported that in 2011, world avocado production reached 4.4 million tons, increasing about 20% from 2007 to 2011; this also increases the quota of waste generated from avocado. Fruit peels, such as avocado peel, are normally not consumed and is consequently rejected. After an investigation on the properties avocado peel, it was noted as a source of nutrients and it was observed to contain phenolic and flavonoid compounds. Due to this investigation, Rotta *et al*. (2016) suggested a tea formulation as a way of reusing the discarded avocado peels. Waste is any material that is discarded, rejected, unwanted or abandoned matter intended for disposing, recycling, re-processing, recovery, re-use, or purification by a separate operation from that which produced the matter, or for sale, whether of any value or not (Lamb *et al*., 2012). Avocado peels are enormous and are about 11 % of the whole fruit, although this proportion vary among cultivars and it must be properly utilized. Extensive research shows that processing agricultural waste (e.g., fruit peels) increases the number of antioxidants compared to the flesh or pulp of the fruits (Rosero *et al*., 2019; Rotta *et al*., 2016; Tian, 2016). Waste has constituted a treat to the existence and well-being of living things on earth. Agricultural wastes vary across different farms and, it is initially not so easy to process except with the right recycling equipment and processing machinery (Industrial Waste Processing and Recovery, 2021). The usual approach to agricultural waste management has been discharge to the environment with or without treatment meanwhile, research has it that fruit residuals have high energy content and bioactive compounds. Extensive research shows that processing agricultural waste (e.g., fruit peels) increases the number of antioxidants compared to the flesh (Rosero *et al*., 2019; Rotta *et al*., 2016; Tian, 2016). Fruit peels or rinds are usually treated as a throwaway but sometimes they could be used as raw materials. One such example is the use of oranges, lemon and pomegranate rinds for preparing face pack and medicines (Fruit Peels, 2021).

Drying is the most commonly used and most energy-intensive unit operation in processing industries, it generally means the removal of water from a material. Drying, a very important method of food preservation is the process in which moisture is vaporized from the surface and within a material, when it comes in contact with air (Keey, 2011). This research is aimed at investigate the effect of air-drying temperature on the drying characteristics, nutritional and bioactive quality of avocado peels.

II. MATERIAL AND METHODS

a. Raw Material and Sample Preparation: Fresh avocados were obtained from a local market in Akure, Ondo State, Nigeria. The ripe avocado fruits were sorted to take out the damaged ones, they were washed with clean water to remove sand and foreign materials. The edible portions and seed of the avocado fruits were carefully separated from the peels. The samples were grouped and kept in a tray wrapped with foil paper in preparation for drying.

b. Chemicals: All the chemicals used were of analytical grade. Ammonium molybdate, Aluminum chloride, Folin Ciocalteu reagent, Gallic acid, Methanol, Sodium hydroxide, Sodium nitrate, Sodium phosphate, Sulfuric acid (H_2SO_4), hydrogen chloride, copper (II) sulphate ($CuSO_4$), ethanol.

2.1 Drying procedure

The drying experiments were performed using a laboratory model oven dryer (DHG-9053A, China) having a constant air velocity of 1.4 ms^{-1}. The dryer consisted of a tray, electrical heater, fan and a temperature controller (40 – 200 °C, dry bulb temperature). Approximately 100g avocado peels were loaded on a metal tray and dried in the oven at 70 °C, 60 °C, 65 °C, 50 °C and 40 °C. Weights of the trays and avocado peels were recorded at intervals of 1 hour during drying, using a digital balance with ±0.01g accuracy until the final moisture content was achieved.

2.2 Nutritional Quality

The nutritional quality investigated include:

a. Moisture content: The initial moisture content of the fresh sample of the avocado peels was determined by oven drying method set at 105 ±2 °C for 72 hours using ASABE S352. standard and applied by; Abodenyi, *et al.*, (2015); Oloyede, *et al*., (2015): Oniya, *et al*., (2016); Olabinjo, (2020). The experiment was replicated and the average weight recorded. The moisture content was evaluated using Equation 1.

$$M.C_{(wb)} = \frac{M_b - M_a}{M_b - M_c} * 100\% \quad (1)$$

where:

MC_{wb} is moisture content (% wet basis),

Mb is the weight of moisture can plus sample weight before oven-drying (g),

Ma is the weight of moisture can plus sample weight after oven-drying (g) and

Mc is weights of moisture can (g).

b. Carbohydrate: The carbohydrate content of the avocado peel was determined by estimation using the arithmetic difference method presented in Equation 2 (Rekha and Rose, 2016; Olabinjo, 2022).

$$\%\text{Carbohydrate} = 100 - (\%\text{ fat} + \%\text{ ash} + \%\text{ fibre} + \%\text{ protein}) \quad 2$$

c. Crude Fibre: it was determined using acid and alkali hydrolysis method as described Oparuku *et al.* (2010); Olabinjo (2020). The ash obtained was cooled in a desiccator and weighed. The percentage crude fibre was calculated using the Equation 3.

$$\% \text{ Crude fibre} = \frac{\text{Loss in weight after drying}}{\text{weight of the sample}} \times 100. \quad 3$$

d. Crude Protein: Protein content of the sample was determined using the Kjeldahl method reported by Rekha and Rose (2016). The total nitrogen was determined and multiplied by a conversion factor of 6.25 to obtain the protein content.

e. **Crude Fat:** It was extracted using AOAC (2012) standard method. About 50 mg of the extracted fat content of the sample was saponified for 5 minutes at 95 ℃ with 3.4 mL 0.5M KOH in dry methanol. The mixture was neutralized using 0.7M HCl. 3 mL of the 14 % boron trifluoride in methanol was added. The mixture was heated for 5 minutes at 90 ºC to achieve complete methylation.

f. Ash content: The ash content was determined using a method reported by Rekha and Rose (2016); Olabinjo (2020). Two grams of the sample was weighed into a crucible in a muffle furnace and heated at 130 °C for three hours until it became gray ash. The weight of ash was obtained by the difference as shown in Equation 4.

$$\%\text{Ash} = \frac{(\text{weight of empty crucible } + \text{ ash}) - \text{weight of empty crucible}}{\text{Weight of sample taken}} \times 100 \quad 4$$

g. Calculated metabolic energy: the metabolic energy in Kcal/100g presented in the sample was determined using Equation 5 as stated by (Akalu and Geleta, 2019). The metabolic energy is measured in Kcal/100g or KJ/100g

$$CME = (9 \times crude\ fat) + (4 \times protein\ conetent) + (4 \times carbohydrate\ content) \quad 5$$

2.3 Bioactive Quality

The bioactive quality investigated in this work include the total phenolic content, flavonoids and vitamin C. The methods used in the determination of these compounds are as follows:

a. Flavonoids: The flavonoid content was determined using a reported protocol by Parsa *et al*. (2017). A clear solution was obtained by taking 0.3 mL extract (0.3 mg/mL in ethanol) and 3.4 mL of aqueous ethanol (30 %) in a test tube. Then 0.15 µL of aqueous sodium nitrite solution (0.5 M) was added followed by 0.15 µL aluminum chloride solution (0.3 M). After a time of 5 min, 1 mL NaOH solution (1 M) was added and the contents were mixed together before measuring its absorbance using UV visible spectrophotometer (UV-1700 Shimadzu) against a blank sample at 506 nm. The blank sample was prepared using the same procedure. In preparing the blank sample an equal volume of methanol was used to replace the plant extract.

b. Total Phenolic Content: Total phenolic contents (TPC) of various extracts were estimated by the method reported by Parsa *et al*. (2017). The sample of each extract was prepared by taking 4.3 mg extract in 10 mL of ethanol and irradiated with ultra-sonics for 5 min to obtain a homogenized solution. Later, 0.3 mL was taken in a test tube and 1 mL methanol; 3.16 mL of distilled water; 0.2 mL of Folin-Ciocalteu reagent were added. The test tube mixture was then incubated for 8 min at room temperature, thereafter 0.6 mL solution (10 %) of sodium carbonate was added in a test tube, covered with aluminum foil and incubated in hot water bath for half an hour at 40 °C. An equal volume of methanol was used to prepare the blank solution by the same procedure used for the preparation of the sample. The absorbance of all the samples was determined using a UV visible spectrophotometer (UV1700 Shimadzu) at 765 nm.

c. Vitamin C: vitamin C, otherwise called ascorbic acid was determined using a method reported by Olabinjo (2020). Thirty grams of the sample blended with reasonable amount of 0.4 % oxalic acid. (4 g/litre) and filtered by What man (No.1) filter paper. The ample volume completed to 250 ml with 0.4 oxalic acid. Twenty ml of filtrate pipetted into a conical flask and titrated with a known strength 2-6-dichlorophenol indophenol until a faint pink colour appeared. The dye strength determined by taking 5 ml oxalic acid 10 % (50 mg/00ml) and added to a standard ascorbic acid (0.05/250ml) oxalic acid 10 % titrated with 2-6-dichloerophenol indophenol (0.2 g/500ml) till faint pink colour expressed in mg/100g (AOAC, 2005).

III. RESULT AND DISCUSSIONS

3.1 Nutritional Composition of Avocado Peel

The results of the nutritional composition from this research were shown in Table 1. The nutritional composition of food materials includes the carbohydrate, crude fibre, crude fat or lipids, protein or amino acid, total sugar, energy content etc. this research investigates the major nutritional compositions of avocado peel. Duarte *et al*., (2016) reported that the avocado pulp contains from 13.5 to 24 % fat, 0.8 to 4.8% carbohydrate, 1.0 to 3.0% protein, 0.8 to 1.5% ash, 1.4 to 3.0% fibre, and energy density between 140 and 228 kcal (Duarte *et al*., 2016). However, the results (Table 1) from this research were found to have higher nutritional content than the pulp (the edible part) reported by Duarte et al., (2016).

a. Moisture content

The initial moisture content of the fresh sample of the avocado peels was determined by oven drying method (AOAC, 2010; Olabinjo, 2020; 2022). The initial moisture content (52.586 %) was reduced to 2.75, 5.82, 6.36, 6.37, 6.64 and 8.06 % wet basis at 70, 65, 60, 50, 40 °C and open sun respectively. It was also observed that at the beginning of the drying process the moisture movement decreased slowly, but at increased drying temperatures the moisture movement decreased rapidly until equilibrium moisture content (EMC) was reached. The Moisture content of the fresh samples was observed to be 52.586 % wet basis which was lower than those recorded by Rotta *et al*. (2016) ($65.05^{a} \pm 3.10$). Increase in drying temperature led a decrease in the moisture content of the sample. The lowest moisture content was recorded for samples dried at 70 ℃ to be about 2.689 %.

b. Crude fat content

Research has it that fats play a vital role in assisting health skin and hair, insulating body organs against shock, regulating body temperature and promoting health cell function (Olabinjo, 2020). Avocado peels had a high crude fat value of 18.988 %, this value is higher than those of the avocado pulp (13.5 – 24 %) as reported by Duarte *et al*. (2016); monkey cola seeds (1.45%) reported by Olabinjo (2020); avocado peel ($12.21^{a} \pm 0.28$) reported by Rotta *et al*. (2016) and papaya peels (2.44 ± 0.25c) reported Santos *et al*. (2014). However, the results from this research are lower than moringa seeds (47.1%) reported by Rekha and Rose (2016) and papaya seeds (29.72 ± 0.37a) reported by Santos *et al*. (2014). The crude fat of avocado peel contributes to the energy value and could be serve as a good source of oil. From the results presented in Table 1, a significant decrease in the value of the fat content of the avocado peels was observed. Sample oven dried at 50℃ (4.916%) had the lowest values for the fat content, which then increased to 6.441% and 7.441% when dried at 60℃ and 70℃ respectively.

c. Protein content

In Nigeria, plant proteins are a major source of food nutrient especially for the less privileged population. Proteins are needed to make life sustaining hormones, important brain chemicals, antibodies, digestive enzymes, and necessary elements for the manufacture of DNA (Olabinjo, 2020). Fresh avocado peels have a value of 4.069 %. The value of protein for fresh avocado peels in this research is slightly lower than protein content of monkey cola seeds (4.42 %), watermelon seeds and moringa seeds (30.9 ± 0.9 and 68.4 %) as reported by Olabinjo (2020) and Rekha and Rose (2016) respectively. Increase in drying temperature led to increase in protein content (at 70, 60, 500, 40 and open sun the value was 9.88 %, 13.027 %, 6.034 %, 11.234 % and 13.829 % respectively). However, samples dried at 50 ℃ (6.034 %) exhibited a significant decrease from 11.234 % (at 40 ℃).

d. Crude fibre content

Fibres helps to clean the digestive tract by removing potential carcinogens from the body and, they also support in the stoppage or absorption of excess cholesterol. Adequate intake of dietary fibre can lower the serum cholesterol level, risk of coronary heart disease, hypertension, constipation, breast cancer, diabetes and colon (Ishida *et al*., 2000 as cited by Olabinjo, 2020). Fibre, is a general word that includes various carbohydrates, hemicelluloses, cellulose, pectin, lignin, gum etc. Fibre enhances other sugars absorption by altering the emptying time of the gastric system and encourages insulin response (Javed *et al*., 2019). The fresh peels of avocado fruit contained crude fibre value of 12.981 % which is higher when compared to moringa seeds (1.2 %) reported by Rekha and Rose (2016), papaya seeds (8.79 ± 0.11c) reported by Santos *et al*. (2014). Avocado peels dried at 60 ℃ (35.187%) had values higher than those of papaya peels (33.05 ± 0.7b) reported by Santos *et al*. (2014). However, avocado peels dried at 70 ℃ (33.294 %) had values in the same range as papaya (33.05 ± 0.7b). From the results in Table 1, a significant increase in the crude fibre content of the avocado peel was observed as the drying temperature was increased. Samples dried at 70 ℃, 60 ℃, 50 ℃, 40 ℃ and under the sun had a crude fibre of 33.294 %, 35.187 %, 30.805 %, 32.182 % and 35.55 % respectively.

e. Ash content

The ash content avocado peels recorded was about 2.298 % for the fresh samples, this value is lower than the value of Rotta *et al*. (2016) avocado peel ($5.43^{a} \pm 0.59$). It was higher than the ash content of monkey cola seeds (1.150 %) as reported by Olabinjo (2020); potato peels (0.6 – 1.6 g/100g) as reported by Javed *et al*. (2019) and lower than the value of ash content of papaya peels and seeds (11.85 ± 0.68a and 6.94 ± 0.79b respectively) as reported by Santos *et al*. (2014); potato peels (6%) as reported by Liang (2014) in Javed *et al*. (2019). However, the dried avocado peels had higher ash content than the values reported by other researchers. The avocado peels dried at 70℃, 60℃, 50℃, and 40℃ and under the sun had an ash content of 5.955%, 5.955%, 6.567%, 6.725% and 6.817% respectively. There was a significant increase between the fresh samples and samples dried at 40℃ which later decreases as the temperature was increased.

f. Carbohydrate content (by difference)

The carbohydrate constitutes a major class of naturally occur organic compound which is essential for maintenance of both plant and animals. The carbohydrate content of fresh avocado peel was found to be 9.078% and was lower than value of monkey cola seeds (26.81%) as reported by Olabinjo (2020); however, peels dried at 70℃ had values (40.732%) higher than those of the avocado o pulp (0.8 – 4.8%) as reported by Duarte *et al*. (2016), papaya seeds and peels (20.73 ± 0.68 and 9.67 ± 1.04b) respectively reported be Santos *et al*. (2014), moringa seeds (25.1 %) reported by Rekha and Rose (2016) and potato peels (8.7 – 12.4 g/100g) reported by (Javed *et al*., 2019). Carbohydrates from plants are one of the three major energy sources in food, along with protein and fat (Olabinjo, 2020). It was observed that an increase in the drying temperature led to an increase in the carbohydrate content of the avocado peel. Samples dried at 70 ℃ (40.732 %) had the highest value of carbohydrate content while those ovens dried at 40 ℃ (29.649 %) had the lowest values. Avocado peel sun dried had a value of about 24.679 %.

g. Calculated metabolic energy

The fresh avocado peels had an energy content of 223.48 Kcal/100g which is lower than the energy content of blanched papaya peel powder was 300 Kcal/100g and blanched papaya peel paste was 43 Kcal/100g as reported by Pavithra *et al*. (2017). The lowest and highest values for avocado peels were recorded for samples dried at 50℃ and 70℃ respectively (230.056 Kcal/100g and 269.417 Kcal/100g respectively). Other drying temperature had values of 235.501 Kcal/100g (60℃), 253.442 Kcal/100g (40℃) and 236.256 Kcal/100g for open sun drying.

3.2 Bioactive Composition of Avocado Peel

The results showed the presence of total phenolic content (TPC) and flavonoids in avocado peel, are accountable for the detected antioxidant properties, owing to their chemical constitution (Rotta *et al*., 2016). Rotta *et al*. (2016) reported that the antioxidant properties of food are widely studied due to their impact on the food quality and the importance of these compounds in maintaining human health. The results of the analysis presented in Table 2 shows that avocado peels are a rich source of bioactive compounds exhibiting antioxidant capacity, similar observations were made by Rosero *et al*. (2019).

a. Vitamin C

Vitamin C is also called ascorbic acid and it is used as an index of the nutrient quality for fruit and vegetable products (Olabinjo, 2020); insufficiency of vitamin C in diets causes the disease called scurvy, which is prevented by as little as 10 mg/day of vitamin C. Vitamin C content of the dried avocado peel ranges from 2.98 mg/100g – 14.303 mg/100g. The fresh avocado peels had vitamin C value of 7.941 mg/100g, the highest value of 14.303 mg/100g was recorded with peels dried at 70℃ followed by 60℃ with value of 13.020 mg/100g and least recorded by oven dried at 40℃ with value of 2.98 mg/100g. The vitamin C content of avocado peels is higher than monkey cola seeds, ranging from 1.108 – 2.229 mg/100g as reported by Olabinjo (2020). Most fruits and vegetables contain vitamin C, which can be reduced by application of heat; however, drying the avocado peels at 70℃ increase the vitamin C value. This proves that the avocado peels are good sources of vitamin C.

b. Flavonoids

Rotta *et al*. (2016) reported that the presence of TPC and flavonoids in avocado peel, are responsible for the antioxidant properties present, owing to their chemical constitution. From the results presented in Table 2, it was observed that increase in drying temperature led to an increase in the flavonoid content of the avocado peels. The fresh avocado peels (3.542 mg/100g) when oven dried at 40°C increased to 16.755 mg/100g. However, when oven dried at 50°C reduced to 16.101 mg/100g; this could be as a result of some unavoidable factors when trying to take the weight of the samples at hourly interval. Sample oven dried at 70 °C and 60 °C had the same value of 21.124 mg/100g; while samples sun dried had values of 21.8 mg/100g. The flavonoid content of the avocado peel oven dried at 60 °C is higher than those of orange peels (3.35 ± 0.603^{A}), lime peels (4.85 ± 0.971^{A}) and lemon peels (8.88 ± 0.621^{B}) dried at 60°C as reported by Erba *et al*. (2020). All values (except those of the fresh samples) recorded in this report was found to be higher than those of dried pomelo peels (4.65±0.02) and bitter orange peels (5.88±0.047) as reported by Pandey *et al*. (2019).

c. Total Phenolic Content (TPC)

Parsa *et al*. (2017) reported that phenolic compounds are usually found in both nonedible and edible plants, which exhibit several biological effects including antioxidant activity. The TPC value of fresh avocado peels recorded in this study was 44.716 mg/100g which was higher than TPC value of monkey cola seeds (14.148 mg/100g) reported by Olabinjo (2020). An increase in drying temperature led to decrease in the value of the TPC; samples dried at 40℃, 50℃, 60℃ and 70℃ had values of about 32.986%, 25.643%, 20.944% and 22.744% respectively. Sun dried samples on the other hand had the least value of about 18.025%. The values of TPC obtained in this research was found to be lower than avocado peel extract (1058.0 ± 59.7g mg GAE/g dried extract) recorded by Rosero *et al*. (2019). However, it was found to be higher than the values pomelo peels (16.09±0.07 mg GAE/g dried extract) and bitter orange peels (6.03±0.05 mg GAE/g dried extract) as reported by Pandey *et al*. (2019).

Table 1: Nutritional Composition of Avocado Peel

S/N	PARAMETER	FRESH	70°C	60°C	50°C	40°C	OPEN SUN
1	Crude fat content (%)	18.988	7.441	6.441	4.916	9.99	9.136
2	Carbohydrate content (by difference) (%)	9.078	40.732	31.356	40.419	29.649	24.678
3	Protein content (%)	4.069	9.88	13.027	6.034	11.234	13.829
4	Moisture content (%)	52.586	2.698	8.034	9.99	10.22	10.489
5	Crude fibre content (%)	12.981	33.294	35.187	30.805	32.182	35.55
6	Ash content (%)	2.298	5.955	5.955	6.567	6.725	6.817
7	Calculated Metabolic Energy	223.48	269.417	235.501	230.056	253.442	236.256

Table 2: Bioactive Composition of Avocado Peel

S/N	SAMPLE	VITAMIN C (mg/100g)	FLAVONOIDS (mg/100g)	TPC (mg/g)
1	Fresh	7.941	3.542	44.716
2	70°C	14.303	21.124	22.744
3	60°C	13.02	21.124	20.944
4	50°C	12.98	16.101	25.643
5	40°C	2.298	16.755	32.986
6	Open sun	9.59	21.8	18.025

IV. CONCLUSION

Fruit peels, such as avocado peel, are not generally consumed and are therefore discarded. After an investigation on the properties, the results showed that avocado peels are rich sources of nutrients and subsequently can be good sources for antioxidants which helps to fight free radicals and cancer. Analysing all the results, it was observed that avocado peel stood out for the following parameters: fibre, ash, phenolic compounds, vitamin C, metabolic energy and carbohydrate. For example, samples dried at 60 °C had values of 35.187 % (fibre), 5.955 % (ash), 20.944 mg/g (phenolic content), 13.02 mg/100g (vitamin C), 235.501 % (metabolic energy), 31.356 % (carbohydrate), 6.441 % (fat), 13.027 % (protein), and 21.124 mg/100g (flavonoids). The knowledge about the nutritional and bioactive composition of avocado peels subjected to drying provides useful information for industries interested in using avocado by-products (the peel flour can be used for dye adsorption), reducing waste, and adding value to the fruit bringing benefits to the environment.

The avocado peels should be dried at mild air-drying temperature between 40 °C and 50 °C to maintain high nutritional and bioactive compounds such as ash, fat, fibre, protein, carbohydrate, metabolic energy, vitamin C, flavonoid and phenolic content. Awareness on the nutritional and bioactive properties of agricultural waste such as avocado peels should be done, to enhance their usage in communities and also to reduce waste.

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Evaluation of Nutritional Composition of Ripe Date Fruit (*Phoenix Dactylifera L.*) pulp and Seed grown in Nigeria

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Abstract— *Date fruit (Phoenix dactylifera L.) is rich in both macro and micronutrients, including vitamins. Several researches have been carried out to determine the nutritional properties of various varieties of date fruit, however, there is currently no study that compares the nutritional composition, total phenolic content and sugar content of ripe red and yellow date. Proximate composition of ripened red and yellow dates was analyzed, the total phenolic content, Vitamin C and sugar contents were also determined. The moisture content in the date samples were 4.87 % and 5.76 %; ash content 1.05 % and 1.08 %; protein content to be 3.11 % and 1.37 %; carbohydrate content to be 60.6 % and 61.02 %; fat content to be 18.02 % and 17.4 % and crude fibre content in the two date samples was determined to be 12.3 % and 13.2 % for Red and Yellow variety respectively. Carbohydrate content of the red date was significantly higher than that of yellow date fruit with both having 14.87% and 9.10% respectively Vitamin C content of red and yellow date was 1.71 mg/100g and 1.46 mg/100g respectively. Vitamin C content of the varieties were fairly low; however, phenolic content was high. The total phenolic content of the red and yellow date was 37.91 mg/100g and 43.55 mg/100g respectively. The finding of this research helps in understanding the nutritional composition of two different varieties of the Nigerian date fruits which can be a basis for developing value-added supplements and nutritious food in the food industry.*
***Keywords*— Date Fruit, Nutritional Composition, fibre.**

I. INTRODUCTION

Date fruit (Phoenix dactylifera L.) is known to be one of the oldest fruit trees in the world (Marzouk and Kassem 2011). They are important subsistence crop grown in hot arid regions but marketed across the world because of its high nutritional and confectionery value (Sanusi *et al.*, 2016). Date tree can tolerate extremely high temperatures and does not wither when cultivated in direct sunlight, thus, it thrives even when grown in the desert (Dada *et al.*, 2012). Date fruits are rich in both macro and micronutrients, including vitamins, consequently, their production and consumption have increased all over the world as a major dietary fruit (Kuras *et al.*, 2020).

In Nigeria, date palm is grown in all the states across the federation, however, it produces and thrives better in the Sudan savanna and guinea vegetation (Sanusi *et al.*, 2016). Thus, production in Northern Nigeria far exceeds production and cultivation in other regions of the nation because of suitable climatic conditions. Its fruit serve as a major food, particularly for breaking Muslim fast, and also serve as an ingredient for making juice, snack and syrups. All parts of the date palm are useful, consequently, they are employed for providing shelter, making timber products and products such as brooms, baskets, mats and ropes (Sanusi *et al.*, 2017).

The fruit's pleasant odour and flavour makes them desirable as food, in addition to their utilization in the beverage and food flavour industry. They are generally known to possess high carbohydrate such as fructose, glucose and sucrose (El-Sohaimy, 2010). Furthermore, they are great sources of dietary fibre and minerals like iron, calcium, potassium and vitamins, but they have low protein and fat content (Agboola and Adejumo, 2013). The flesh of dates contains between 60-65 % sugar, 2 % protein, about 2.5 % fibre, and less than 2 % each of minerals, fat, and pectin substances (Zaid *et al.*, 2002). Its sugar content is essential for individuals who cannot tolerate sucrose.

Dates contain numerous phytochemicals such as carotenoids and phenolics, which contribute to the fruit's antioxidant activity. Therefore, they possess good, antiviral, antimutagenic and anticancer activities (Assirey, 2015). The concentration of the phytochemicals decreases as ripening or maturity occurs. Its rich hydroxyl pope folic acid makes it fit for increasing the body's immunity and resistance to cancers (Ali *et al.*, 2012). All these properties sum up to make date fruit essential in providing strength, boosting immunity and fitness, providing relief during pain and protection against diseases such as cancer and other heart related diseases.

Date fruit is considered an ideal food because of its broad spectrum of usefulness. However, a large percentage of the Nigerian populace, including the date palm industry are still oblivious of its benefit despite its tremendous nutritional and medicinal potential. Therefore, the objective of the research was to determine the nutritional composition of two varieties of date fruit cultivated in Nigeria with respect to the proximate, vitamins and antioxidant composition. The research will also help to provide data of the fruit useful for food reformulation and developing value-added supplements and food in the food industry.

II. MATERIALS AND METHODS

2.1 Sample preparation

Two ripe varieties of date fruit (Red and Yellow) in the research were procured from the main market, Zaria, Kaduna state, Nigeria. The samples were cleaned and washed before any experiment was done.

2.2 Chemical analysis

All the samples were analyzed chemically using the official methods described by the Association of Official Analytical Chemist (AOAC, 2010). Moisture content of each samples used in the experiment was determined by taking measurement of the sample before and after water was removed by the process of evaporation. The samples were placed in the oven and dried for 8hrs at 105℃. The gravimetric method described by AOAC (2010) was adopted in determining the percentage of ash in the samples. The fat content was determined using the Soxhlet type of the direct solvent extraction method. Micro-kjedhal method was used in determined the protein content of the samples.

2.3 Determination of Carbohydrate content

The total carbohydrate content of each sample was estimanted by "difference". The sum of the percentage concentrations of each parameter of the other proximate compositions were subtracted from 100. The total carbohydrate content was calculated as equation 1 by Olabinjo (2020; 2022);

Total carbohydrate = 100 - (% moisture + % ash + % fat + % protein + % crude fibre) …… (1)

2.4 Determination of Total Phenolic Content

The total phenol content of the sample was determined by the method of (L. Fu, (2010). 0.20 ml of the plant extract was mixed with 2.5 ml of 10% Folin ciocalteau's reagent and 2.0 ml of 7.50 % sodium carbonate solution. The reaction was mixture was subsequently incubated at 45 ℃ for 40mins and the absorbance of the coloured mixture was read at 700nm using UV visible spectrophotometer. Garlic acid was used as standard phenol.

2.5 Determination of Vitamin C Content

The vitamin C content was determined using the ascorbic acid as the reference compound. 200 ul of the extract was pipetted and mixed with 300 ul of 13.30 % of TCA and 75ul of DNPH. The mixture was incubated at 37 ℃ for 3hours and 500 ul of 65 % H_2SO4 was added. The absorbance was then read at 520nm (AOAC, 2006).

2.6 Statistical Analysis

The data obtained from the experiment were analyzed using the statistical package for social science software (SPSS version 16.0). Analysis of variance (ANOVA) test was used in determining the mean ± standard error, while Duncan's multiple range test was used in separating the means at significant level of p = 0.05.

Fig.1; Bulk of the two ripe varieties of date fruit (Red date fruit and Yellow date fruit)

III. RESULTS AND DISCUSSION

3.1 Proximate composition of the banana varieties

Proximate composition of the ripe date fruit pulp and seeds were investigated to understand the nutrient profile of the fruit and its quantity in terms percentage. The proximate analysis revealed the presence of ash, moisture, fat, carbohydrates, fibre and protein in the two varieties of date fruit (*Phoenix dactylifera*) pulp and seed. Table 1 shows the mean proximate composition of the ripe varieties of Date fruit (red and yellow) pulp and seed used for the study. The result indicated that the amount of nutrients in each of the samples varied.

Table 1: Table showing the mean proximate composition of ripe red and yellow date fruit pulp/seed (g/100 g).

Sample	Ash	Moisture	Fat	Fibre	Protein	CHO
A	1.23 ± 0.004^a	59.70 ± 0.09^e	2.24 ± 0.04^b	19.80 ± 0.12^d	2.17 ± 0.01^b	14.87 ± 0.05^c
B	0.64 ± 0.003^a	67.11 ± 0.04^f	4.17 ± 0.12^c	16.77 ± 0.05^e	2.23 ± 0.03^d	9.10 ± 0.11^b
C	1.05 ± 0.004^a	4.87 ± 0.06^e	18.02 ± 0.06^b	12.30 ± 0.12^d	3.11 ± 0.01^b	60.63 ± 0.05^c
D	1.09 ± 0.003^a	5.77 ± 0.04^f	17.48 ± 0.12^c	13.27 ± 0.05^e	1.38 ± 0.03^d	61.02 ± 0.11^b

Notes: A: Ripe Red Date Fruit pulp, B: Ripe yellow date fruit pulp; C: Ripe Red Date Fruit seed, D: Ripe yellow date fruit pulp. Results in the table above are presented as mean ± standard deviation.

*Values followed by different letters in each column are statistically different at $p < 0.05$.

i. Ash content: - From table 1, the yellow date fruit variety had an ash content of 0.64 %, while the ash content of the red variety was 1.23 % and the ash content of the Red date seed variety is 1.05 % and 1.08 % for Yellow variety. The red Date fruit pulp had more ash content than the yellow pulp variety while reverse was the result for the date fruit seed. This value was lower than those obtained by Habib and Ibrahim, (2011) and Abdulrahman *et al.*, (2020). The ash content in the study showed the percentage of inorganic mineral elements present in both varieties date fruits. Generally, ash content is considered as an index of mineral present in a food or index to the nutritive value of foods (Agboola and Adejumo, 2013). Thus, the moderate level of ash content shows a moderate level of minerals present in the date fruits. This result review that the samples have low ash content when compare to 3.27 % by Ogungbenle (2011). This result of both varieties is lower than 1.6% of kimri as reported by Gamal et al. (2012). This result is in agreement with the result obtained from the analysis of different varieties of date palm fruits (1.3 for Dora), (1.7 Dhaki) and (1.5 Karbaline) respectively (Faqir et al., 2012).

ii. Moisture content: - The Ripe red date pulp had a moisture content of 59.70 %. while that of ripe yellow date was 67.11 %. The moisture content of both pulp variety was higher than those reported by Assirey (2015) and Abdulrahman *et al.,* (2020), whose result showed that moisture content of a selected number of date fruits ranged between (10.5 - 29.5 g/100 g dry weight) and (2.25 – 7.65 %) respectively. The moisture content recorded for the seed were lower in value compared to the pulp. This variation can be attributed to differences in variety, stage of maturation, environmental conditions (Biglari, 2009). High moisture content in dates fosters spoilage, while low moisture content will make the fruit not unacceptable to consumers. The result revealed that the moisture content in the two date seeds were 4.87 % for Red variety and 5.76 for Yellow variety. This result shows that the moisture content of Yellow variety is higher than that of the Red date variety. The moisture content value recorded in this research were lower compared to different varieties of date palm seeds, (7.81 % Dora), (9.90 for Dhaki), (6.3 for Karbaline) respectively as reported by Faqir *et al*., (2012).

iii. Fat content: - The fat contents for red and yellow date fruit pulps were 2.24 and 4.17 % respectively. The date seed containing a significant amount of fat ranged between 17.4 % for Yellow variety and 18.02 % for Red variety, showing that the red date seeds had more fat content than the yellow variety. The low-fat content of red and yellow dates pulp indicates that they are safe for high blood pressure patients as they contain low levels of cholesterol and fatty acids. This was within range of that reported by Abdulrahman *et al.,* (2020). However, the fat content is higher in values as reported by Assirey (2015), (Habib and Ibrahim, 2011) for Saudi Arabian, Iranian, and United Arab Emirates varieties, which ranged between (0.12-0.72%), (0.4–0.9%), and (0.10-0.21 %) respectively. The results reported in this work were more than those reported by El-Rahman (2017), who found that date seeds contained high levels of crude oil, being 5.95 and 6.4 times as high as that in date fruits of palm shell, respectively. This percentage favors the extraction of seed oil, which has many benefits. The study of two Tunisian cultivars by Besbes *et al.* (2004) showed that date seed oils contain high relative percentages of oleic acid. They are also more yellow-colored than other vegetable oils and they can protect against UV light responsible for much cellular damage. Date seed oils could easily be conserved due to their high oxidative stability. Regarding these specifics, the value of this by-product in the cosmetic and food industries may be justified. However, the antioxidant composition of date seed oil must be tested in order to more valorize this byproduct.

iv. Fibre content: - Fibre content of the red date pulp was 19.80 %, while that of the yellow date was 16.7 7%. The crude fibre content of seeds were 13.2 % for Yellow variety and 12.3 % for Red variety as presented in Table 1. Dates are excellent sources of dietary fibre, which has tremendous health benefits. It is consumed to prevent the incidence of heart disease, colon cancer, diabetes and other disorders (Habib and Ibrahim, 2011). Furthermore, the moderate quantity of fibre in the date fruits makes them suitable for aiding absorption and digestion process when consumed. Fibre content of foods helps in digestion process and prevention of cancer (Saldanha, 1995; UICC/WHO, 2005). Crude fiber decreases the absorption of cholesterol from the gut in addition to delaying the digestion and conversion of starch to simple sugars, an important factor in the management of diabetes (Cust et al., 2009). Dietary fiber serves as a useful tool in the control of oxidative processes in food products and as functional food ingredient (Mandalari et al., 2010).

v. Protein content: - Red date fruit pulp had a protein content of 2.17 %, which was slightly lower than that of yellow date fruit pulp. The yellow date seeds variety had a protein content of about 1.37 % while the red date seeds variety had protein content of about 3.11 % (Table 1). Dates generally have been reported to have low protein content; thus, they are not a good source of protein (Sirisena *et al.*, 2015). These results were lower than those of Saudi date variety (3.12 %) as reported by El-Rahman and Al-Mulhemsi (2017). The results also showed that the red date seeds had more protein content than the yellow date seeds variety.

vi. Carbohydrate content: - Carbohydrate content of the red date fruit pulp was significantly higher than that of yellow date fruit pulp with both having 14.87 % and 9.10 % respectively. Dates are generally good sources of carbohydrates and fibre. This result indicates that the ripe red and yellow date fruit pulp and seeds are great energy sources because of their high carbohydrate and fibre content. More so, it shows that the ripe red variety is suitable for confectionary products. The results presented in table 1 shows that the yellow date seeds had more carbohydrate content than the Red variety. The value of carbohydrate present in the two varieties are 60.6 % for Red variety, and 61.02 % for Yellow variety. This value is lower than 80.67 % (Dhaki) as obtained Ogungbenle (2011).

3.2 Total phenolic contents of the Date fruit varieties

Total phenolic content, which is an antioxidant compounds in plants, plays a great role in fighting diseases in humans and promoting healthy living. They are known to possess antibacterial, antiviral and anti-inflammatory diseases.

Date fruit analysis show that the content of antioxidant compounds is dependent on the variety and place of cultivation. (Kuras *et al.,* 2020). Phenolic content was considered an effective antioxidant because they act as free radical captors or scavengers. Several research groups have reported that dates are rich in phenolic acids (Benmeddour Z. *et al.*,2013; El Sohaimy S. *et al.,* 2015). The total phenolic content of the red and yellow date pulp recorded in the study were 37.91 mg/100g and 43.55 mg/100g respectively (Table 2). This shows that yellow date fruit pulp has more phenolic content than red date fruit pulp. Red variety of date seed contained the highest total phenolic content (31.4 mg/g), whereas Yellow variety had the lowest total phenolic (31.2 mg/g). The variation in the phenolic content can be attributed to the heterogeneous nature of Date fruit in terms of variety, place of cultivation, growing condition, maturity, season, geographic origin, fertilizer, soil type, storage condition, and amount of sunlight received (Kuras *et al.,* 2020). Both varieties fruit are recommended as a major diet for treating gastric disease conditions and other therapeutic purposes because of their high phenolic content. The results of phenolic contents were higher than those reported by El-Rahman and Al-Mulhemsi (2017), who found 10.3mg/ g in varieties grown in Oman. Seeds may be considered a rich source of phenolic compounds based on high phenolic content. Date seeds could be used in functional foods, food additives, pharmaceuticals, and cosmetic industries.

Table 2: Table showing the Total phenolic content, vitamin C and sugar content of the ripe Date fruit pulp and seed for the yellow and red varieties.

Sample	A	B	C	D
Phenolic content (mg/100g)	37.91	43.55	31.41	31.29
Vitamin C (mg/100g)	1.79	1.46	NA	NA
Sugar content (mg/100g)	0.24	0.23	NA	NA

3.3 Vitamin C contents of the Date fruit varieties

Date fruits provides a broad range of essential nutrients; hence, they possess potential health benefits upon consumption. The results from table 3 showed that both variety of dates were in the same range of Vitamin C content with red date having 1.71 mg/100g and yellow date having 1.46 mg/100g. Vitamins present in the fruits show that they can aid multiple biochemical reactions in organisms when digested.

3.4 Sugar contents of the Date fruit varieties

The result also showed that both varieties have similar sugar content; 0.24 and 0.23 mg/100g respectively for red and yellow dates. Glucose, fructose and sucrose are some of the sugars found in dates. Although analysis of these individual constituents was not carried out, the result as tabulated on table 2 shows that both ripe red and ripe yellow date fruits have low sugar contents. Sugars are one of the most important constituents of dates, the low sugar content of the two varieties show that they can be consumed by diabetic and high-blood pressure patients.

IV. CONCLUSION

This study shows the nutritional composition of two ripe varieties of date fruit (Phoenix dactylifera l.) cultivated in Nigeria with respect to the proximate, vitamins and antioxidant composition. The varieties used for this experiment (red and yellow variety) contained substantial nutrient levels which varied in both the ripe varieties. The study also shows the heterogeneous nature of date fruit with respect to variety, and place of cultivation. Both varieties of the dates are excellent sources of carbohydrate. The rich fibre content of the varieties makes them suitable for aiding absorption and digestion process when consumed. Vitamin content of the varieties were fairly low; however, phenolic content was high. Sugars are one of the most important constituents of dates, however, the low sugar content of the two varieties show that they can be consumed by diabetic and high-blood pressure patients. Further researches can be done to determine the mineral, and antinutrient properties of different varieties of Nigerian date fruit.

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Sedimentation dynamics of soil particles in sylvopastoral half-moons on restored plateaus in the Ouallam department (Niger)

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Abstract— *The present study was conducted on the Tondibiya and Satara plateaus, in the Ouallam department (western Niger). The objective is to study the sedimentation dynamics of soil particles deposited in sylvopastoral half-moons of different years (2014, 2016 and 2018). The assessment of sedimentation was done in selected half-moons following a survey on transects. It involved the determination of the thickness of the sedimented horizon, the measurement of the permeability by the single ring infiltrometer and the granulometric analysis in the laboratory. The results obtained showed that the thickness of the sedimented horizon varies according to age and is 14.15, 17.33 and 19.37 cm in the 3, 5 and 7 year old half-moons respectively. Water infiltration was optimised by sedimentation. It was recorded 70 minutes to be constant in the 7-year half-moon and 40 minutes in the 3-year half-moon. The granulometric analysis of the sedimented particles shows their dominance by sands (77.73%). These different results show that the sylvopastoral half-moon is effective in improving the physical parameters of the soil on degraded ferruginous plateaus.*

Keywords— Restoration, Soil, Sedimentation, Ouallam, West Niger.

I. INTRODUCTION

In Niger in general, and in its western part in particular, natural resources are undergoing degradation due to the combined actions of climate and man (Boni *et al*., 2016; Abdou *et al*., 2016). This situation has been accentuated since the droughts of the 1970s and 1980s (Larwanou, 2005). Each year, more than 250,000 ha of arable land are washed away by degradation (GEF-IFAD, 2002). The dynamics of degradation are mainly reflected in the significant retreat of natural vegetation formations (tiger bushes and steppes) in favor of developed landscapes and denuded soils (Issoufou *et al*., 2018). The consequences include a continued decline in agricultural and pastoral production at a time when basic needs are increasing. Increasingly, ecosystem services are being disrupted, significantly impacting the socio-economic conditions of local populations, especially during periods of climate shocks (Douma, 2016).

In view of the increasing degradation of natural resources, the consequences of which contribute to the development of the phenomenon of desertification (Larwanou, 2005), a dynamic of restoration of degraded lands is developing nowadays. Indeed, to restore degraded lands, techniques of Water and Soil Conservation / Soil Defence and Restoration (CES/DRS) have been commonly carried out in Niger. Based on the typology of the landscape unit to be managed, these different techniques have been designed according to the desired goal. Among these CES/DRS techniques, dug structures, including the half-moon for silvopastoral purposes, are the most widely used (Douma *et al.*, 2011; Amani *et al.*, 2021). However, apart from the respect of design and management standards, the results of the developed areas depend on the age of the construction, but also vary according to the sites (Vlaar, 1992; Laminou *et al*., 2020). It therefore appeared necessary to study the impact of the sylvopastoral half-moon on the physical conditions of the soil. Thus, the objective of this work is to evaluate the physical parameters of sediments deposited in the half-moon on pastoral sites developed in different years.

The department of Ouallam in western Niger was the study area (Fig. 1). The developed plateaus of Tondibiya and Satara, in the rural communes of Tondikiwindi and Simiri respectively, were the study sites for the present work. The climate is tropical arid, with an average annual rainfall of 250-450 mm (Laminou *et al.*, 2020). The year is characterised by a dry season of 8 to 9 months and a rainy season of 3 to 4 months. The average minimum temperature is around 18° (December-January) and the maximum around 45° (March-April). This rise in temperature leads to an increase in evapotranspiration, which is around 3000 mm per year, with a minimum of 460 mm in the rainy season and 2460 mm in the dry season (Amadou,2012).

II. MATERIALS AND METHODS

2.1. Study sites

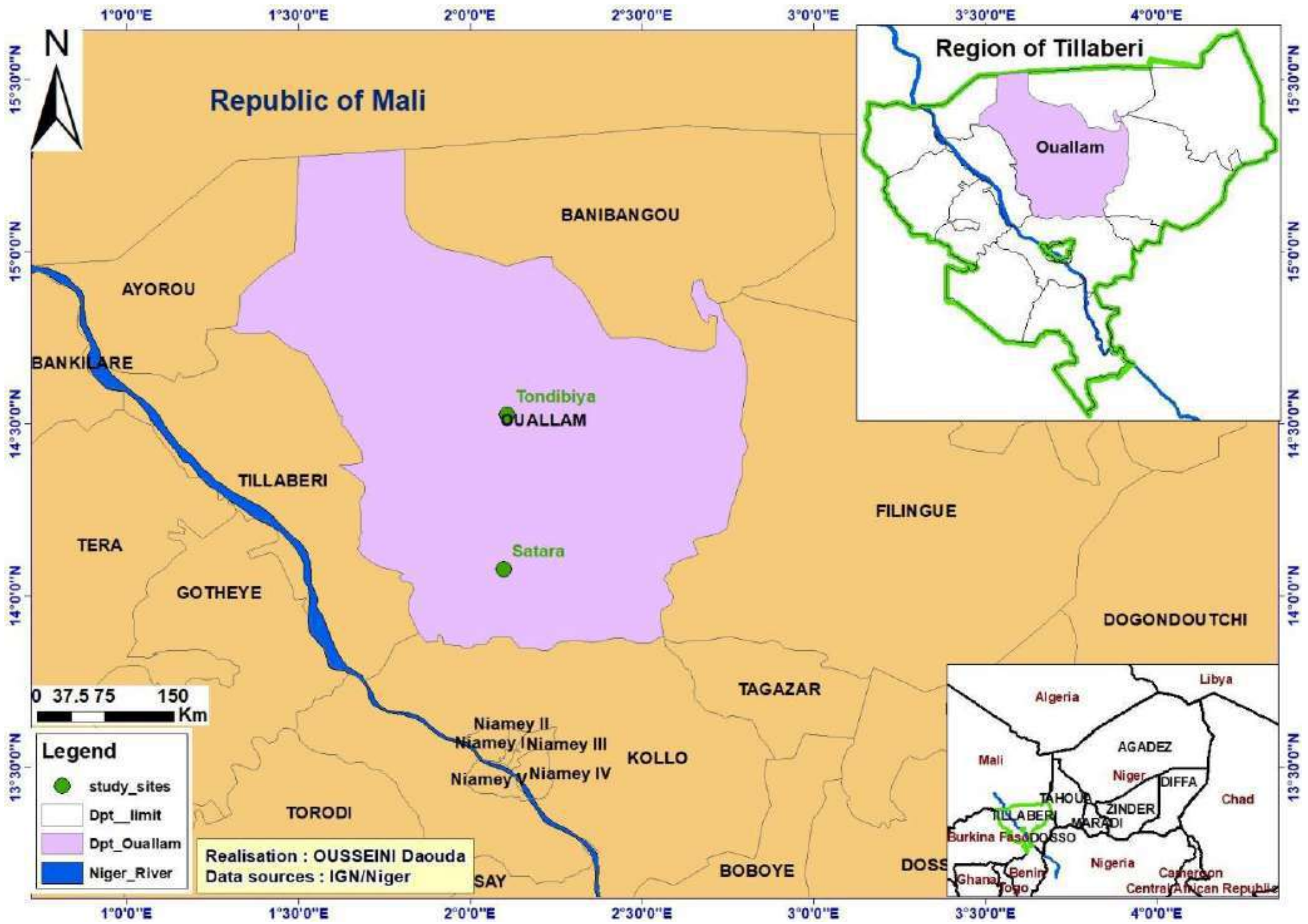

Fig. 1: Map of Department of Ouallam

The sylvopastoral half-moon, the subject of this study, has the characteristics presented in Table 1. Photo 1 illustrates a half-moon.

Table 1: Characteristics of a half-moon

Characteristics of the Half-moon sylvopastoral pastoral	**Values**
Diameter (m)	4
Depth of the trough (m)	0,15 à 0,30
Bead height (m)	0,30 à 0,40
Area (m^2)	32
Density per hectare	313

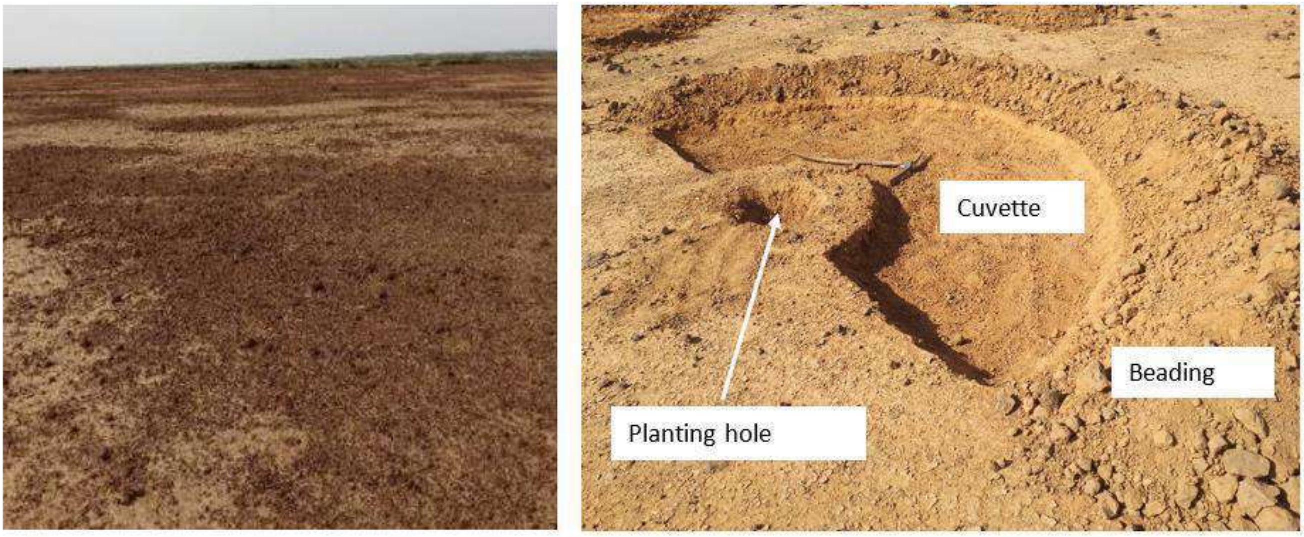

Photo 1: a: View of an undeveloped site; b: Compartment of a completed half-moon

The biological treatment given to the structures when they were installed and the natural regenerative power of the environment have allowed vegetation to return. The sites were constructed in different years starting in 2014. Thus, the study selected the 2014, 2016 and 2018 completion sites. Data collection was carried out in 2020 where the respective ages of these sites are 7; 5 and 3 years.

2.2. Data collection

The data was collected mainly on the particles deposited in the half-moon basin. This sedimentation assessment was done along three (3) parallel transects. One transect runs perpendicular to the contour line. On each transect, ten (10) half-moons were selected using the "No sounding" method according to formula (1). The number of half-moons surveyed per site was thirty (30). The sampling by transects and sampling were chosen to have the representativeness of the structures taking into account the factors influencing the dynamics of sedimentation among which the wind (Abdourhamane, 2011).

$$\boldsymbol{P} = \frac{\boldsymbol{N}}{\boldsymbol{10}} \qquad \textbf{P}: \text{No survey} \qquad \textbf{(1)}$$

N = Total number of DLSP on the transect : ∑Xi

Indeed, the deposition of the particles was studied on three parameters: thickness, water permeability and texture. Fig. 2 shows the data collection points in the half-moon.

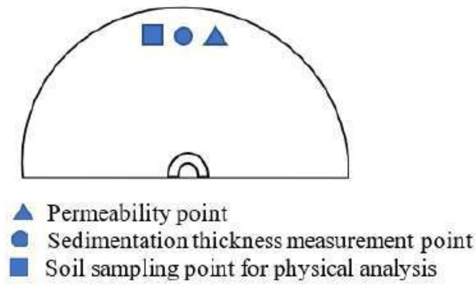

Fig. 2: Collection points in the half-moon

2.2.1. *Thickness of sedimentation*

The thickness of the sediment deposited in the half-moon basin was measured by opening a cultivation profile (Photo 2). The layer representing the deposited soil particles was delimited by means of a visual assessment. The thickness was then measured with a ruler.

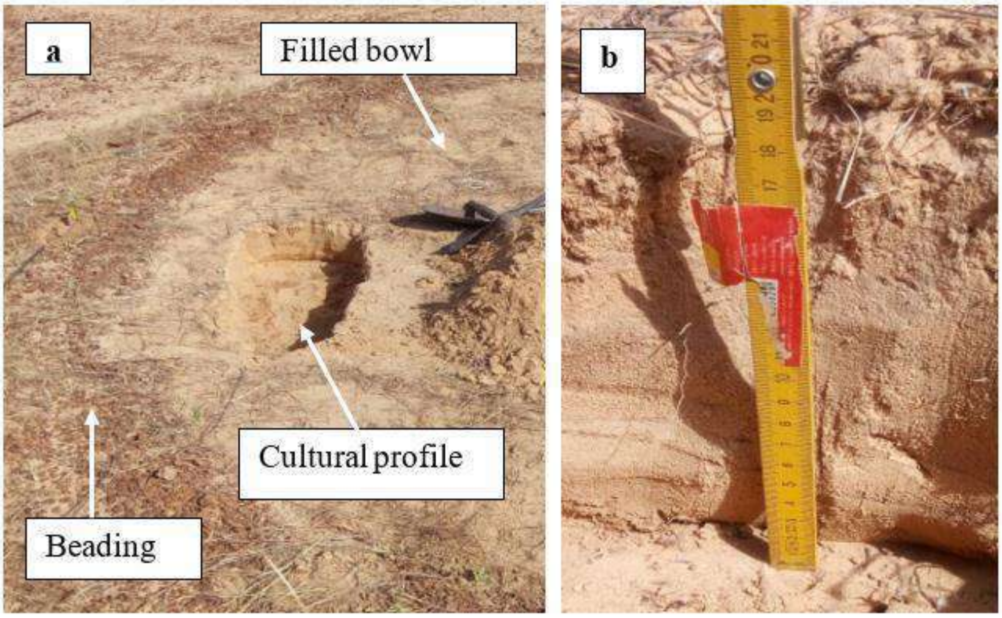

Photo 2: a: Cultural profile in a half-moon filled basin; b: Measurement of deposition

The thickness of the sediment layer depends on the age of the structure. This led to the evaluation of the sedimentation rate as given by formula (2).

$$Vs = \frac{E}{A} \qquad (2)$$

Vs : Sedimentation rate (cm/year); E: Measured sedimentation thickness (cm) and A: Age of the structure in years (year)

The evaluation of the thickness of the sedimentation thus makes it possible to assess the volume of soil available to the herbaceous plants for the development of their subterranean part on the one hand, and available to the water for its infiltration and evaporation flows on the other.

2.2.2. *Water permeability*

The 20 cm diameter single ring infiltrometer was the equipment used to measure water permeability. The activity consisted of installing the infiltrometer and quantifying the infiltrated water depths. The infiltrometer was sunk about 5 cm into the ground to prevent water from leaking onto the ground surface during the test. It is then filled with potable water to a height (H) and allowed to infiltrate for a timed period. At the end of the time, a height (Hf) is obtained from which the infiltrated height (Hi) is obtained by formula (3). The operation is repeated by filling the infiltrometer again to the height H. The activity is thus continued repeatedly until a constant infiltration height is obtained (same Hi value three times in a row). The number of repetitions of the operations during a test (Table 2), depends on the time taken to obtain Hi. The infiltration rate or infiltration coefficient (Yaméogo *et al*., 2013) was calculated according to formula(4).

$$\mathbf{Hi = H - Hf}$$

Hi : Infiltrated height in millimeters (mm); **H** : Initial height in millimeters (mm); **Hf** : Final height in millimeters (mm)

$$V = \frac{Hi}{t} \qquad (4)$$

V: Infiltration speed mm/min; **Hi:** Inf height in millimeters (mm); **t:** Time taken in minut)

Table 2: Duration of the survey sheet operations

Filling-infiltration operation	Duration in minutes (min)	Number of repetitions
1	5	6
2	10	6
3	20	3

The infiltration test was carried out in the pit of the structure and on a bare area without vegetation representing the control (Photo 3). The permeability allows the sedimented soil to be compared to the encrusted control, as insufficient water infiltration into the soil is a key factor in land degradation or fodder production on pastureland.

Photo 3: Installation of the infiltrometer on an unfinished control

2.2.3. Analysis of textural elements

The physical analysis of the soil particles concerned the first 10 cm in the basin and an undeveloped area representing the control. Sampling was carried out across a crop profile (Photo 4).

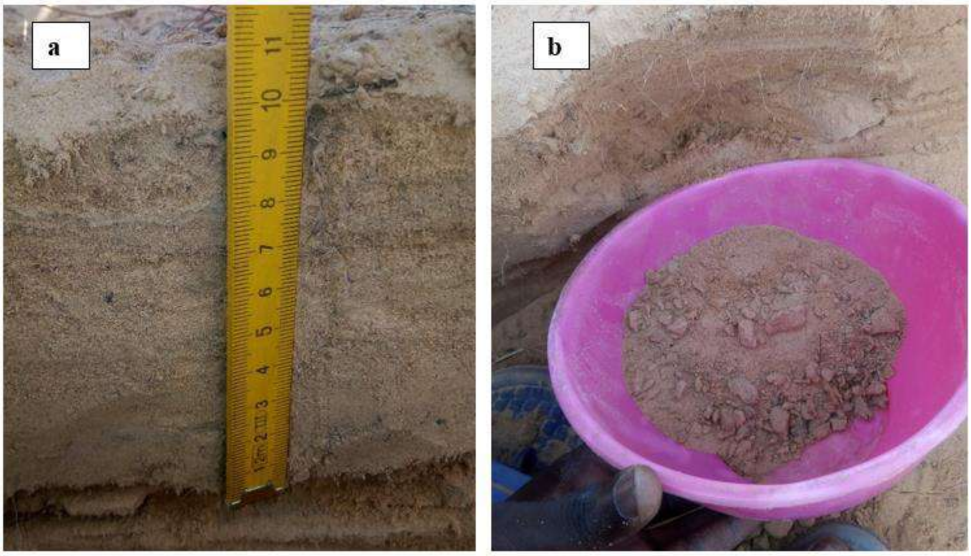

Photo 4: a: Core measurement, b: Soil sampling

A composite sample was taken per transect. Soil samples were analysed in the soil laboratory of the Faculty of Agronomy of Abdou Moumouni University of Niamey. The Robinson Pipette method was used for the particle size analysis. The textural elements concerned are clays, silts and sands. The textural class was determined using the textural triangle.

III. RESULTS

3.1. Thickness of sedimentation

Table 3 shows the sediment layer thickness values at the different sites.

Table 3: Thickness and sedimentation rate as a function of the age of the half-moon

Age of structure (years)	Deposit thickness (cm)	Sedimentation rate (cm/year)
7	19,37 ± 6,04	2,23 ± 1
5	17,33 ± 1,79	3,46 ± 0,35
3	14,15 ± 1,50	5,85 ±1,80

The results of the sedimentation thickness assessment show that the deposition of particles increases with age. At the 7 year old site, the half-moons are mostly filled in. This has allowed herbaceous species to colonise the basin (Photo 5).

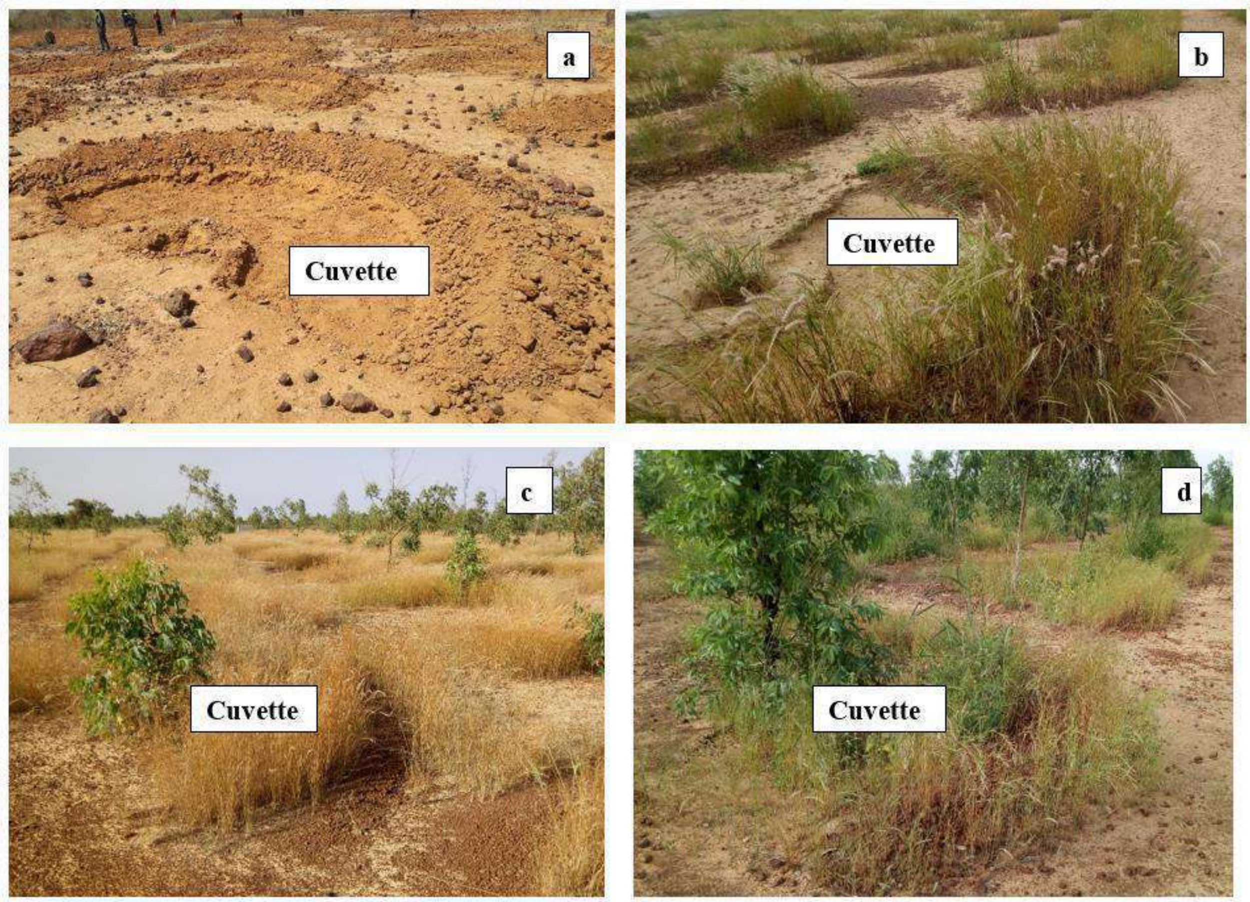

Photo 5: a: Half moons achieved; b: Half moons of 2 years; c: Half moons of 7 years

The sedimentation of soil particles in the half-moon basin has improved the vegetation cover with increasing age. This has an impact on forage production by improving species diversity but also by increasing the amount of biomass.

3.2. Water permeability

The water infiltrates more into the sedimented soil of the basin than into the control. This is due to the coarse-textured granulometry and the porosity found. The cumulative infiltration heights and the infiltration time are different depending on the site. Table 4 gives the cumulative infiltration heights prior to constant infiltration. For the time, 70 minutes were recorded for constant infiltration in the 7-year basin and 40 minutes for those of 5 and 3 years.

Table 4: Cumulative heights at constant infiltration

Measuring points	Cuvette		Witness	
Age of Half moons (years)	Hc (cm)	Tc (mn)	Ht (cm)	Tt (mn)
7	8	70	1	10
5	22,4	40	4,7	15
3	27,9	40	57,2	50

Hc: Cumulative infiltration heights before constant infiltration in the basin; Tc: Duration from which infiltration was constant in the basin, Ht: Cumulative infiltration heights before constant infiltration on the control and Tt: Duration from which the infiltration was constant on the control.

It can be seen that the volume infiltrated and the time taken to obtain constant infiltration on the sedimented soil are greater than those of the control, with the exception of the 3 year old structure. The permeability study shows that infiltration and its speed increase with the age of the structure (Fig. 3 and 4).

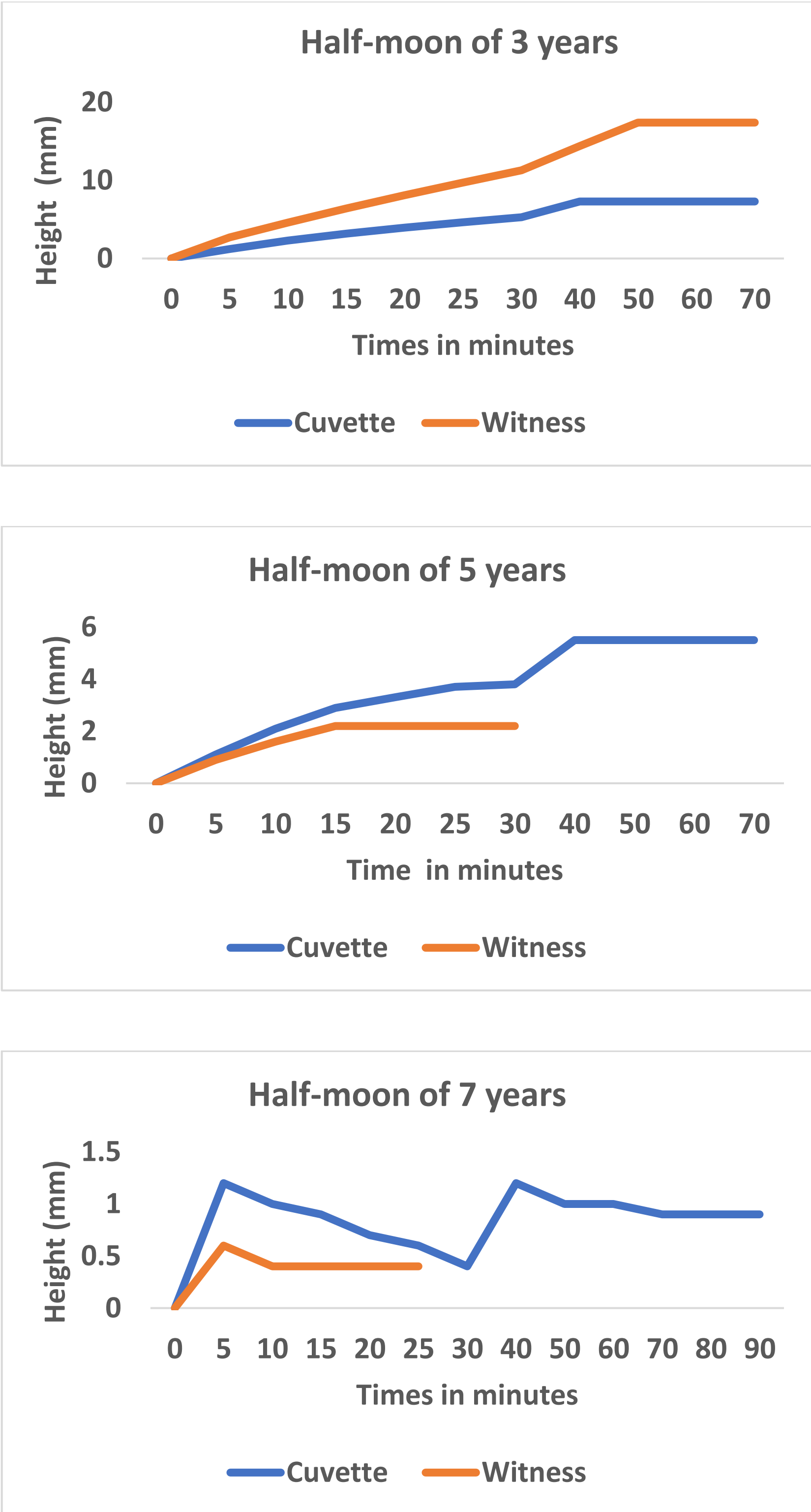

Fig. 3: Evolution of water infiltration in the half-moon

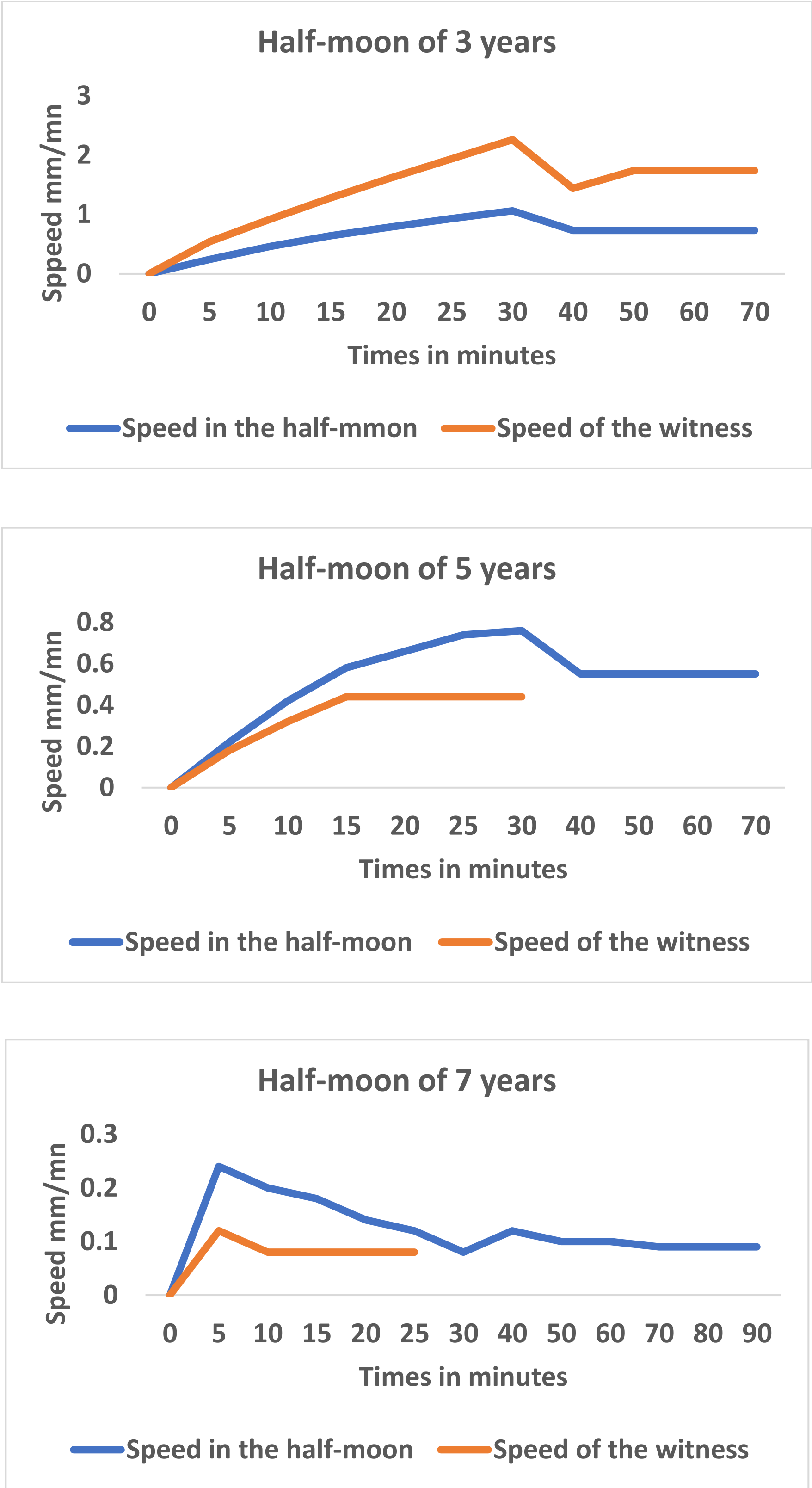

Fig. 4: Evolution of the water infiltration rate

It can be seen that infiltration has been improved by the half-moon. The better infiltration recorded on the 3 year old control site is explained by the fact that the upper soil horizon has more pores than the sedimented layer in the half-moon.

3.3. Particle size parameters

Table 5 presents the results of the particle size analysis.

Table 5: Soil particle size

Half-moon age (years)	Clay	Fine silts	Coarse silts	Fine sands	Medium sands	Coarse sands	sands	Clay + Fine silts	Texture
7	3,78	9,13	9,37	26,26	41,78	9,69	77,73	22,28	Sandy-silt
5	2,52	15,03	12,87	21,96	36,00	11,61	69,57	30,42	Silty-sandy
3	1,01	12,36	11,93	30,37	33,71	10,61	74,69	25,30	Sandy-silt
TCD	3,04	12,09	15,36	21,77	33,21	14,52	69,5	30,49	Silty-sandy

Clay ϕ < 2 µm; Fine silts: 2 < ϕ< 20 µm; Coarse silts 20 < ϕ< 50 µm; Fine sands: 50 < ϕ < 200; Medium sands 200 <ϕ< 500 µm and Coarse sands 500 < ϕ < 2000µm and TCD: Completely degraded control

The granulometric analysis reveals that sands dominate. They represent at least 2/3 of the particles with a dominance of medium sands. It also appears that the texture of the sedimented soil in the basin of the structure tends to be coarse. The 7-year-old half-moon recorded more sands compared to the 5- and 3-year-old half-moons.

IV. DISCUSSION

The deposition of particles in the basin has the role of reconstituting the essential functions of soils as a support for species, particularly plant species, as a nutrient bank and as a catalyst for exchange between the edaphic environment and the atmosphere (Ay *et al*., 2020). The thickness of the sedimentation varies between sites, but the trend shows that it increases with the age of the half-moon. It increases from 14.15±1.5 cm in the 3-year-old half-moon to 19.37±6.04 cm in the 7-year-old half-moon. Apart from age, factors influencing the deposition of particles in the basin can include wind erosion, crumbling of the bead by raindrops, the texture of the developed soil and the type of development works. In their study in the commune of Simiri (Niger), Laminou *et al.* (2020) found a sedimentation thickness of between 8 and 15 cm in the 3-year-old pastoral bench, the structure sharing the same basic principles and functioning with the pastoral half-moon. This proves that excavated structures cause sedimentation of particles in their trough.

Depending on the site, the infiltration of water, the height of infiltration, the time from which it becomes constant and the speed of infiltration are different. This difference tends to become apparent as the age of the structure increases. In this sense, the duration from which the infiltration becomes constant is 40 and 70 minutes respectively for the 3 and 7 year old half-moon. This is explained by the thickness of the sedimentation which increases with age. The high infiltration rate or infiltration coefficient of the structures is 1.06 mm/min, obtained in the basin of the 3-year old half-moon. Laminou *et al.* (2020) found an average infiltration rate of 1 cm/min (10 mm/min). This difference may be due to the heterogeneity of the plateau horizons and the age of the structure. In Burkina, Yaméogo *et al.* (2013) obtained an infiltration coefficient of 0.1.10-3 m/s (6.25 mm/min) and 0.04.10-3 m/s (2.5 mm/min) in the zaï, respectively for the basin and the control. These results confirm that infiltration depends on the type of development techniques, the nature of the soil and also the age of the structure. The various results show that the excavated structures significantly improve the permeability of water in the soil.

Indeed, the improvement of the permeability in the half-moon following the deposition of particles, leads to the improvement of the humidity in the structure. Thus, Kagambega *et al.* (2011) found that the development of the half-moon significantly influenced the humidity rate both in the middle of the rainy season (August) and at the beginning of the dry season (end of October) in Burkina Faso. In their work on water balance modelling (Burkina Faso), Zouré *et al*. (2019) stated that half-moons are able to mitigate the effect of droughts by keeping water available for plants over extended periods of up to three weeks. This ability of the half-moon was possible due to the infiltration and availability of water in the basin. Meanwhile, in southwest Niger, Douma *et al.* (2011) found that an estimated 7% improvement in moisture was achieved in the first year of the silvopastoral trenches. They added that the moisture could further increase in the future with the age of the structures due to progressive sedimentation. This highlights the decisive character of time in the production of ecosystem services, illustrating a dynamic in the structures on the restored sites.

The evolution of the silty-sandy texture of the completely degraded control (TCD) to that of the sandy-silty texture of the 7-year-old half-moon implies that the accumulation of sands has taken place. This constitutes an improvement of the structure, of which the herbaceous flora is the main beneficiary, given that encrustation remains the main cause of the degradation of the plateaus. The age and the structure are factors determining the textural dynamics in the structures. Development therefore favors the deposition of sand (André *et al*., 2008) and the development of herbaceous plants due to their strong resilience to the Sahelian climate. In their study, Amani *et al.* (2021) found that the creation of the bench induced a change from medium to fine texture dominated by clays. This points to the fact that the excavated works of degraded land reclamation techniques contribute to the evolution of the texture on the degraded ferruginous plateaus of western Niger, dominated by medium and low or non-sandy textures (CILSS, 2016). In parallel to the developed plateaus, Tidjani (2013) obtained on a stabilised dune flat in the department of Gouré (Niger), 91.11% of sands for the A horizon (0 to 25 cm). These results show that the sediments deposited would be related to the nature of the soil and the type of development on the one hand and confirm that the techniques of recovery of degraded land make the texture of the soil evolve on the other hand. Land reclamation techniques have always improved the physical characteristics of soils (Hamado, 2011; Abdou *et al*., 2020). Despite their low physico-chemical potential, reclaimed degraded soils offer very appreciable fodder production (Kiema *et al*., 2012).

V. CONCLUSION

At the end of this study, it was found that the half-moon allowed the deposition of soil particles. The thickness of the sediments increases with the age of the structure. After seven years, the basin is completely filled in. This sedimentation provides a support for vegetation, particularly herbaceous vegetation, which has developed. We note the evolution of the texture which becomes coarse thanks to the accumulation of sand in the structure. This has led to an improvement in the infiltration of water into the soil. The result is an improvement in humidity, which creates a microclimate at the scale of the half-moon. As with all techniques for reclaiming degraded land, the sylvopastoral half-moon creates edaphic conditions for the development of herbaceous species, especially grasses, which have a high ecological resilience in the Sahel. This implies that the plateaus are suitable for sylvopastoral recovery.

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Risk Control Model of Paddy Rice Farming Production and Farmers' Behavior in Tanjung Jabung Barat (Moscardi and De Janvry Approach Method)

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***Abstract*— *The role of rice farming in meeting rice needs is linear with the behavior of farmers responding to production risk. Farmer behavior determines the magnitude of production risk and farmer decision-making. The behavior of farmers is risk averse, risk neutral, or risk taker. Assessing the actual production function, the frontier production function, the risk function, and farmer behavior are helpful as benchmarks to make it easier to answer what factors determine the level of efficiency and use of optimal production input scenarios to achieve optimal production. The purpose of this study is (1) to analyze the response and production risk response to the use of production inputs. (2) Analyzing the behavior of farmers in responding to production risks. (3) Building a model for handling production risks and farmer behavior. The research was conducted in Tanjabbar Regency which was determined proportionally, the sample size was based on the slovin method as many as 122 farmers, and the sampling method was simple random sampling. The method of data analysis is the Cobb-Douglass production function, the Cobb-Douglass risk function, the behavior of farmers using Moscardi and De Janvry, and the production risk management model and the behavior of farmers using the kumbakar function. Cultivation technology carried out by farmers is still conventional with the use of production inputs that are still below the recommended dose. The productivity obtained by farmers is low, and the production risk is high. The determinants of the productivity function of lowland rice farming are urea, SP36, KCL, and organic fertilizers. Optimal use of these production inputs will be able to reduce the occurrence of production risk. Farmers' behavior in responding to production risks is to avoid risk and is mainly determined by the demand for urea and SP36 fertilizers. Sources of technical inefficiency mostly come from a narrow land area. Production risk control can be done by increasing productivity through the use of optimal production inputs, especially urea, SP36, and organic fertilizers, as well as the intensification of land area.**

***Keywords*— Response, Production Risk, Farmer Behavior, Optimal Production.**

I. INTRODUCTION

Nationally, rice farming is cultivated by approximately 18 million farmers and contributes 66 percent to the gross domestic product (GDP) of food crops. Not only that, rice farming has provided income and employment opportunities for the total household, which exceeds 21 million with a contribution of 25 to 35% of its income. Therefore, rice continues to be a strategic commodity in food security and the national economy, as well as being the main basis for future agricultural revitalization. Significantly, national rice production has increased almost three times from 1970 to 2020. This condition is certainly related to the increase in planted area and productivity. The increase in rice productivity reached 87.6% in the period 1970 to 2020 with a productivity of 5.56 tons/ha in 2020, while for 1970 it was 2.42 tons/ha. The area has increased during this period by 39.8 percent. In 1970 the harvested area was 8.3 million hectares and became 11.6 million hectares in 2019. Nationally, the increase in rice production cannot be separated from government policies in the fields of intensification, extensification, and policies for agricultural machinery assistance, particularly government policy support, institutional engineering, technology cultivation, as well as superior varieties (Research and Development Center, 2021).

Jambi Province is ranked 18th as a national rice producer. Harvested area in 2014 was 121,722 ha and became 140,992 ha in 2020 or an increase of 15.83%. Production in 2014 was 587,384 tons and became 729,424 tons in 2020 or an increase of 24.18% per year. Productivity in 2014 was 48.26 quintal/ha and became 51.74 quintal/ha in 2020 or an increase of 7.2% per year. For Jambi Province, the Tanjabbar Region is the third place as the center for rice production. In 2014 the harvested area of lowland rice was 8,403 ha, and production was 32,730 tons with productivity of 38.95 quintal/ha. In 2020 there was an increase so that the harvested area became 9,569 ha, production of 50,118 tons with the productivity of 52.37 quintal/ha for Tanjabbar Regency, the center of rice production is Batang Asam District during the period 2014-2020 harvested area of 2,850 ha with a production of 19,730 tons with productivity 5.8 tons/ha.

The role of rice farming in meeting rice needs always goes hand in hand with the behavior of farmers in responding to production risks. Farmer behavior determines the magnitude of production risk or is related to decision-making. There is a difference between farmers who are afraid of risk and those who like risk. If farmers are risk averse, they will use fewer inputs and be careful about using inputs. Meanwhile, farmers who are risk-takers will use more inputs. (Pujiharto, 2017).

Farmers' decision-making takes the risk of being influenced by the behavior of farmers and affects the demand for production inputs. The amount of use of production inputs will affect the allocation and application of production inputs. Farmers in farming need to make decisions regarding risk-taking whether they are risk takers, risk-averse, or risk-neutral. In this regard, facing production risk is determined by the farmer's response (behavior) to production risk-taking attitudes and depends on the courage to take risks. If the courage of farmers is greater in facing risks, the use of production inputs in farming will be even greater. In this regard, production risk is determined by the degree of behavior and response of farmers in responding to production risks and depends on the level of response of farmers in the use of production factors.

II. RESEARCH METHODS

This research was conducted in Tanjabbar Regency with the research locus of Batang Asam District as the production center. The research area took two villages which were carried out purposively with the consideration that these locations had the potential for the development of rice farming. The research locus was Rawa Medang village and Sri Agung village. The sample size used the slovin method with a precision level of 10%. From a farmer population of 1,820 households, the number of samples was 122 households. The sampling method uses Simple Random Sampling with a random table.

Data analysis method

The analysis of the Cobb-Douglas production function, both the actual production function and the frontier production function, refer to Soekartawi (2006) and Tasman, A (2008).

The form of the Cobb-Douglas production function to analyze the actual production function is the Ordinary Least Square (MOLS) method.

$$\mathbf{Y_0 = \beta_0\ X^{b1}}$$

$$\mathbf{Y = b_0 X1^{b1} X2^{b2} X3^{b3} X4^{b4} X5^{b5} X6^{b6} X7^{b7} X8^{b8} X9^{b9} e^{u}}$$

$$Ln\ Y = Ln\ a + b1 Ln\ X1 + b2 Ln\ X2 + \cdots + bn Ln\ Xn + V$$

The form of the Cobb-Douglas production function to analyze the frontier production function is the Maximum Likelihood Estimation (MLE) method.

Mathematically the stochastic frontier function is significant in the following equation:

$$\mathbf{Yi = x1\beta + (Vi - Ui);\ where\ i = 1,2,3,\ldots N}$$

The form of the transformation of the stochastic frontier function is significant as follows:

$$LnY = \beta_0 + \beta 1 LnX1i + \beta 2 LnX2i + \beta 3 LnX3i + \beta 4 Lnb4i + \beta 5 LnX5i + \beta 6 Lnb6i + \beta 7 LnX7i + \beta 8 LnX8i + \beta 9 LnX9i + (vi - ui)$$

The general form of the function is transformed by the equation notation as follows:

$$\mathbf{ln\ PRO = b_0 + b_1\ lnLT + b_2\ lnBE + b_3\ lnPN + b_4\ lnPP + b_5\ lnPK + b_6\ lnPO + b_7\ lnPT + b_8\ lnPJA + (vi - ui)}$$

Where :

PRO = Production Produced by Farmers (kg)

b0 = Constant

LT = Use of Planted Area (ha)

BE = Seed Use (kg)

PN = fertilizer use N (kg)

PP = Use of Fertilizer P (kg)

PK = Use of Fertilizer K (kg)

PO = Use of organic fertilizer (kg)

PT = Use of drugs (ml)

PJA = use of labor (HOK)

b1-b8 = Estimating Parameters For Variable LT...PJ

A u = Error

E = 2.718 (Natural logarithm)

Test the hypothesis using the value (p-value),

- p.value > α (0.05), > α (0.01); Ho accepted
- p-value < α (0.05), < α (0.01) ; Ho rejected

Cobb-Douglas Risk Production Function Analysis

The CD production function model of the risk function is as follows:

$$LnY\ \sigma 2=(Yi-\hat{y}i)^2 = Ln + \alpha 1 LnX1i + \alpha 2 LnX2i + \alpha 3 LnX3i + \alpha 4 Lnb4i + \alpha 5 LnX5i + \alpha 6 LnX6i$$

$$+ \alpha 7 LnX7i + \alpha 8 LnX8i + \alpha 9 LnX9i + \varepsilon$$

Yi : production of paddy rice farming

Ŷ : Frontier production of paddy rice farming

The general form is transformed into the application model as follows:

ln PRO* = b0* + b1* lnLT* + b2* lnBE* + b3* lnPN* + b4* lnPP* + b5* lnPK* + b6* lnPO* + b7* lnPT* + b8* lnPJA*

PRO* = Production Produced by Farmers (kg)

b0*= Constant

LT*= Use of Planted Area (ha)

BE*= Seed Usage (kg)

PN* = fertilizer use N (kg)

PP* = Fertilizer Use P (kg)

PK* = Use of Fertilizer K (kg)

PO*= Organic Fertilizer Use (kg)

PT*= Drug use (ml)

PJA* = use of labor (HOK)

b1*-b8*= Estimating Parameters For Variable LT*...PJA*

Farmer Behavior Measurement

The method used to measure the behavior of farmers using the Moscardi and de Janvry method with the model:

There are three categories of classification of farmer behavior towards risk, namely:

$$K(s) = \frac{1}{\theta}\left(1 - \frac{PxiX}{Pyfi\mu y}\right)$$

Where:

θ= Coefficient of variation of production (θ = Va / Ea) ,where Va = Standard deviasion and Ea = Average production of rice farming

Py= Price of Product

fi= Elasticity input to production

X_i= The number of factors of production – i

Px_i= Price of factors of production – i

μy= Production average

K(s)= Estimated parameter (Parameter) of the behavior of farmers facing the risk

There are three categories of classification of farmer behavior towards risk, namely:

1. Low risk category if risk taker (likes risk) with (0 < K(s) < 0.4)
2. Moderate risk if risk neutral (risk-neutral) with (0.4 < K(s) < 1.2)
3. High category risk if risk averse (avoiding risk) with (1,2 < K(s) < 2.0)

Production Risk Control Model and Farmer Behavior

Pujiharto and Wahyuni (2017), Production risk control models and farmer behavior are formulated from the optimal production function. The optimal production function is formulated from the optimal production input model by Tasman, A (2008) and Soekartawi (2012) with the following econometric equation;

ln PRO = b0** + b1** lnLT** + b2** lnBE** + b3** lnPN** + b4** lnPP** + b5** lnPK** + b6** lnPO** + b7** lnPT** + b8** lnPJA****

Where:

PRO** = Production Produced by Farmers (kg)

b0**= Constant

LT**= Use of Planted Area (ha)

BE**= Seed Usage (kg)

PN** = fertilizer use N (kg)

PP** = Fertilizer Use P (kg)

PK** = Use of Fertilizer K (kg)

PO**= Organic Fertilizer Use (kg)

PT**= Drug use (ml)

PJA** = use of labor (HOK)

b1**-b8**= Estimating Parameters For Variable LT**...PJA**

The optimal use of production inputs for controlling production risk and farmer behavior is;

Land Area : 1* = (-FX1)/ *

So that, : $X1 = -\frac{\beta 1^{*} \pi^{*}}{Fx1}$

Seeds : 2* = (-FX2)/ *

So that, : $X2 = -\frac{\beta 2^{*} \pi^{*}}{Fx2}$

Urea : 3* = (-FuX3)/ *

So that, : $X3 = -\frac{\beta 3^{*} \pi^{*}}{Fux3}$

SP_{36} : 4* = (-Fsp36X4)/ *

So that, : $X4 = -\frac{\beta 4^{*} \pi^{*}}{Fsp36x4}$

KCL : 5* = (-F_{KCL}X5)/ *

So that, : $X5 = -\frac{\beta 5^{*} \pi^{*}}{FKCLx4}$

Labor : 6* = (-WX6)/ *

So that, : $X6 = -\frac{\beta 6^{*} \pi^{*}}{FWx6}$

Organic Fertilizer : 7* = (-pX7)/ *

So that, : $X7 = -\frac{\beta 7^{*} \pi^{*}}{Fpx7}$

Chemical Insecticide : 8* = (-pX8)/ *

So that, $X8 = -\frac{\beta 8^{*} \pi^{*}}{Fpx8}$

III. RESULTS AND DISCUSSION

Analysis of the Productivity Function of Lowland Rice Farming

The input variables used in farming will be described and analyzed in the frontier production function model. Productivity analysis aims to determine the variables that affect the productivity of lowland rice farming in the research area. The results of the estimation of the production function can be seen in Table 1.

Table 1. Results of Estimating the Productivity Function of Paddy Rice Farming in the Research Area, 2022

Variable	Productivity Function			
	Coefficient	**Std.Error**	**T-Statistic**	**Prob.**
X1_SEEDS	0,1935	0,0546	3,5439	0,0000
X2_UREA	0,3742	0,0858	4,3613	0,0000
X3_SP36	0,615	0,0367	16,757	0,0003
X4_KCl	0,0546	0,0356	1,533	0,0435
X5_ORGANIC	0,1093	0,0812	\1,3460	0,0261
X6_DRUGS	0,1267	0,0548	2,3120	0,0333
X7_LABOR	0,1856	0,0452	4,106	0,0000
R-squared	0,834352			
Adjusted R-squared	0,854862			

Table 1 shows that the model is free from the classical assumption test. The Durbin Watson stat value is 1.605998, which means the model is free from autocorrelation, meaning that there is no correlation between the residuals in one observation and other observations in the regression model. The value of Adjusted R-Squared = 0.863924 means that 86.39% of rice production variables (output) can be explained together by production input variables (seeds, urea fertilizer, SP36 fertilizer, KCl fertilizer, organic fertilizer, drugs, and labor). while the remaining 13.61% is determined by other factors outside the model. The coefficient value of the variable productivity of seeds, urea fertilizer, SP36 fertilizer, KCl fertilizer, organic fertilizer, medicine and labor were respectively 0.397494; 0.124915; -0.024508; 0.080550; 0.127330; -0.042369; 0.297411. Variable seeds, urea fertilizer, SP36 fertilizer, KCl fertilizer, organic fertilizer, medicine, and labor are added by 10% with the assumption that ceteris paribus will increase the productivity of each by 3.97%; 1.24%; -0.24%; 0.80%; 1.27%; -0.42%; 2.97%.

The value of $\sum\beta_1$ = 0.960823 < 1, means that each additional use of production factors by 10% will increase productivity by 9.60%. The scale of lowland rice farming in area II is Decreasing Return to Scale, meaning that each addition of the same proportion of production inputs will result in a decrease in additional production output. **The seed variable (X1)** has a very significant effect. on productivity assuming the use of other inputs ceteris paribus. Consistent with Sutawati's research (2014) regarding the use of seeds that are still not optimal and have a significant effect on the output of lowland rice farming. **The variable urea fertilizer (X2)** has a very significant effect on productivity with the assumption that the use of other inputs is ceteris paribus. This is in line with Damayanti's research (2014) that the use of urea fertilizer has a positive and very significant effect on the productivity of lowland rice farming. **The SP36 fertilizer variable (X3)** has no significant effect on increasing productivity assuming the use of other inputs ceteris paribus. Consistent with Nurani's research (2014) that the addition of SP36 fertilizer will reduce the productivity of lowland rice farming. **KCl fertilizer variable (X4)** has a significant effect on increasing productivity assuming the use of other inputs ceteris paribus. Consistent with research by Sutawati (2014) that increasing rice productivity can be done by increasing the use of KCl fertilizer. **The organic fertilizer variable (X5)** has a very significant effect on increasing productivity with the assumption of using other inputs ceteris paribus. Consistent with research Firmana (2016) that the coefficient value of organic fertilizer is positive, which means that the use of organic fertilizers can increase the value of the efficiency of lowland rice farming techniques, and Conscience (2014) that the addition of organic fertilizer use will increase productivity paddy rice farming. **The drug variable (X6)** has no significant effect on increasing productivity with the assumption that the use of other inputs is ceteris paribus. In contrast to Wulandari (2017) that drugs are positive and have a very significant effect on lowland rice productivity. **The labor variable** has a very significant effect on increasing productivity with the assumption that the use of other inputs is ceteris paribus. Consistent with the research of Nainggolan et al (2016) that the use of labor in lowland rice farming has a significant effect on production.

Analysis of the Technical Efficiency of Lowland Rice Farming Using the Stochastic Frontier Approach

The value of technical efficiency is categorized as quite efficient if ET > 0.7 and categorized as not efficient if ET ≤ 0.7. Coelli (1998) in Silitonga (2016) that the value of the technical efficiency index analysis results is categorized as efficient if it produces a value > 0.7 as the efficiency limit. Farmers who have a technical efficiency index below 0.7 can be targeted for counseling on improving farming management and agricultural techniques. The results of the analysis of technical efficiency in lowland rice farming can be seen in Table 2.

Table 2.Technical Efficiency in Lowland Rice Farming in the Research Area, 2022

Technical Efficiency	Number of Farmers (Person)	Percentage (%)
0.40 – <0.50	12	9,83
0.50 – <0.60	27	22,13
0.60 – <0.70	46	37,70
0.70 – <0.80	29	23,77
0.80 – <0.90	8	6,55
Total	**122**	**100**
Lowest Technical Efficiency	**0.48**	
Highest Technical Efficiency	**0.87**	
Average Technical Efficiency	**0.66**	

Source: Primary Data Results, 2022

Table 2 shows that the average ET = 0.66, meaning that the average productivity achieved by lowland rice farmers is around 66% of the frontier production. In order to achieve efficiency in the use of inputs in the production process, it is necessary to increase as much as 34%. The average lowland rice farming is still not technically efficient. The results of the technical efficiency analysis also show that the lowest technical efficiency for rice farmers is 0.48 and the highest is 0.87. That is, the opportunity to increase production by 13 - 52 percent. Consistent with research by Nainggolan et al (2019) that 40.23% of farmers have ET values > 0.7. Consistent with Febriansyah (2019) that the level of efficiency is low <0.7, which means that farmers have not optimized the use of production inputs properly and have not used them as recommended.

Rice Farming Production Risk Analysis

The Just and Pope production risk function model equation consists of a production function and a production variance function. The most commonly used functional format in the framework of the Just and Pope production risk model is the Cobb-Douglas function in natural logarithm form. The production risk of lowland rice farming is known in the use of lowland rice production factors and can be analyzed using the Just and Pope production risk function model.

Table 3. Results of Estimating the Risk Function of Rice Field Farming in the Research Area, 2022

Variable	**Risk Function**			
X	**Koefisien**	**Std.error.**	**T.statistic**	**Prob.**
X1_SEEDS	0,1258	0,0851	0,1478	0,0426
X2_UREA	-0,3654	0,1436	2,5446	0,0047
X3_SP36	-0,4362	0,1054	4,1385	0,0014
X4_KCl	-0,1534	0,0712	2,1545	0,0412
X5_ORGANIC	-0,0562	0,0293	1.9181	0,0183
X6_DRUGS	-0,0815	0,0541	1,5065	0,0346
X7_LABOR	-0,1252	0,0417	3,0024	0,0053
X8_LAND_AREA	0,0765	0,0323	2,3684	0,0167
R-squared	0,794556			
Adjusted R-squared	0,811544			

Table 3, shows that the production risk function model provides a coefficient of determination (Adjusted R-squared) of 0.8113 This shows that 81.10% of the diversity of lowland rice production risks can be explained simultaneously by factors of seed production, urea fertilizer, fertilizer SP36, KCl fertilizer, organic fertilizer, liquid insecticide, labor, and land area. The results of the analysis obtained Fcount of 349.54 with Prob. 0.0000 which means that the independent variables present in the model simultaneously have a significant effect on the risk of lowland rice production. Prob value. < α (0.05) shows the results that have a significant effect, meaning that the independent variables contained in the model jointly affect the risk of lowland rice production. **The seed variable (X1)** has a negative and significant effect on production risk. That is, the seed variable is a risk-reducing factor. Consistent with the research of Suharyanto (2015) that the production risk regarding the use of seeds has no significant effect on reducing the risk of lowland rice farming production. **The variable urea fertilizer (X2)** has a negative and significant effect on the risk of lowland rice production. That is, the variable urea fertilizer is a risk-reducing factor. Consistent with Apriana (2015) it is significant that the chemical fertilizer variable is a production variable that can reduce production risk. **The SP36 fertilizer variable (X3)** is positive and has no significant effect on the risk of lowland rice production. That is, SP 36 fertilizer is a risk-increasing factor. The results of this study are not in line with the research of Malik et al (2019) that the SP36 fertilizer variable is a variable that has a negative and significant effect on production risk. **The KCl fertilizer variable (X4)** is positive and has a significant effect on the risk of lowland rice production. This means that the KCl fertilizer variable is a risk-increasing factor. Consistent with Apriana's (2015) research, it is significant that the chemical fertilizer variable is a production variable that can reduce production risk. **The organic fertilizer variable (X5)** is positive and has a significant effect on the risk of lowland rice production. That is, organic fertilizer is a risk-increasing factor. The results are different from Apriana (2015) that the chemical fertilizer variable is a production variable that can reduce production risk. **The drug variable (X6)** is negative and has a significant effect on the risk of lowland rice production, meaning that the drug variable is a risk-reducing factor. Consistent with Puspitasari (2011) who indicates that pesticides are a risk-reducing factor. **The labor variable (X7)** is positive and has a significant effect on the risk of lowland rice production. That is, labor is a risk-increasing factor. Consistent with Tahir (2011), it is significant that labor will increase production risk. **The variable land area (X8)** is positive and has a significant effect on the risk of lowland rice production. That is, land area is a risk-reducing factor. Apriana (2015), that land area has a very significant effect on the risk of lowland rice production. The results are different from Apriana (2015) that land input is an input that is a risk-increasing factor.

Farmers' Behavior Against Risk

In this study, the measurement of farmers' behavior towards risk uses the Moscardi and de Janvry method. The Moscardi and de Janvry method is a risk measurement that is carried out by selecting the most significant variable that determines the level of farmer behavior in avoiding risk. The equation of the Moscardi and de Janvry method uses the Cobb-Douglas production function based on the production function, production variations, product prices, and factors of production.

To determine farmers' behavior towards risk or K(s) value, namely the production function. The most significant inputs and the greatest coefficients are selected as inputs of urea and SP36 fertilizers. Urea fertilizer has a probability of 0.0000 and SP36 has a coefficient of 0.131570. Therefore, the variables urea fertilizer and SP36 fertilizer are used as parameters for determining the category of farmer behavior towards the risk of lowland rice farming. Farmer's behavior towards risk can be seen in Table 4.

Table 4. Farmers' Behavior Against Risks in Research Areas, 2022

Urea Fertilizer Variable		
Risk Behavior Criteria	**Frequency (Farmers)**	**Percentage(%)**
Risk Taker	5	4.09
Risk Neutral	41	33.60
Risk Averse	76	62.29
Total	**122**	**100.0**
SP_{36} Fertilizer Variable		
Risk Behavior Criteria	**Frequency (Farmers)**	**Percentage (%)**
Risk Taker	4	
Risk Neutral	28	
Risk Averse	90	
Total	**122**	**100,0**

Table 4 shows that most of the farmers who use the urea fertilizer variable as a parameter for determining behavior with the criteria of avoiding risk (risk averse) which is 78.72 percent and while farmers using SP36 fertilizer as a parameter for determining behavior with the criteria of all farmers avoiding risk (risk aversion). averse). Lowland rice farmers' aversion to risk is motivated by the use of production inputs. Farmers have a tendency to risk-averse behavior causing the allocation of production inputs below the optimum level so that in the end it will result in a low level of productivity. Capital in farming is still very limited, especially for farmers who control narrow lands causing the level of technology adoption to be low which results in low farm productivity.

Low productivity faced by farmers affects the desire of farmers in carrying out their production activities, if there is an increase in the price of production inputs, the demand and use of production inputs will decrease. Lack of farming capital will also lead to a lack of input, which creates production risks and causes farmers to be afraid of risk. Increased productivity affects the desire of farmers to carry out their production activities, if there is an increase in production input prices, the demand and use of farmers' production inputs will decrease. Lack of capital Farming will also cause a lack of input, which creates production risk. Ermelinda's research (2019) shows that all respondents are risk averse. The result of determining farmer's behavior towards risk with urea fertilizer variable. Ermelinda (2011), that all farmers are risk averse. By using the classification of farmer behavior towards risk according to Moscardi and de Janvry, it can be concluded that most of the lowland rice farmers in the study area have risk-averse behavior in the category of risk (1.2 < K(s) < 2.0).

The Influence of Socio-Economic Factors on Technical Inefficiency of Paddy Rice Farming

There are many factors that influence the failure to achieve technical efficiency in the production process, one of which is socio-economic factors. Therefore, the analysis of the sources of technical inefficiency aims to answer what are the causes of technical inefficiency. The sources of technical inefficiency can be seen in Table 5.

Table 5. Result of Estimated Sources of Technical Inefficiency

Variable	Coefficient	Std. Error	t-Statistic	Prob.

Z1	0.1487	0.4052	0.36697	0.0007
Z2	-0.1514	0.4376	-0.34597	0.0002
Z3	-0.0451	0.4011	-0.11244	0.1675
Z4	-0.1145	0.6321	-0.18114	0.0433
Z5	-0,.0676	0.0213	3.17370	0.0048
C	0.4876	0.2247	2.17000	0.0051

Table 5 shows that the value of Adj R2 = 0.578721, this means that 57.87% of technical inefficiency can be explained jointly by socio-economic factors, while 42.13% is influenced by factors outside the model. Variables that have a significant effect on technical inefficiency in lowland rice farming in the study area at the level of α = 0.05 are the variables of age, farming experience, and the distance between the land and the farmer's house. While the variables that are not significantly different from technical inefficiency are education and the number of family members.

The estimation results of the age variable (Z1) have a positive and significant effect on technical inefficiency. The results of the significance test obtained the value of Prob. 0.0003 < α (0.05) which means that it is significantly different. Regression coefficient β 1 = 0.0872 which means that the age elasticity is positive for technical inefficiency, in other words, the age of the farmer has a significant effect on increasing the technical inefficiency of rice farming. This is in line with the increasing age of farmers, the ability to work, the desire to take risks, and the desire to carry out new innovations which are decreasing so that the level of technical efficiency decreases. Consistent with Rika's research (2017), the age of farmers significantly affects technical efficiency.

The education variable (Z2) has a negative and no significant effect on technical inefficiency. The results of the significance test obtained Prob. 0.1730 > α (0.05) which means the difference is not significant. Regression coefficient β 2 = -0.1001 which means that the elasticity of education is negative for technical inefficiency, in other words, a change in the level of education results in a decrease in the level of technical efficiency. The influence of education in the study was due to the fact that the farmers had long experience in farming lowland rice so the farmers worked on farming based on previous experience so that farming would increase and be technically efficient. Consistent with Fauziyah (2010) which has a significant negative effect on education and technical efficiency of farming.

The farming experience variable (Z3) has a negative and significant effect on technical inefficiency. The results of the significance test obtained the value of Prob. 0.0266 < α (0.05) which means significantly different. The regression coefficient β 3 = -1.974 means that the elasticity of farming experience is negative for technical inefficiency, a change in the increase in the level of farming experience will result in a decreasing level of inefficiency. More farming experience can allocate the use of farm productivity inputs better because it will allocate the use of inputs based on previous farming experience so that farming is more technically efficient. Consistent with Firmana (2016) which has a significant positive and significant effect on farming experience to increase the technical efficiency of lowland rice farming.

The variable number of family members (Z4) is negative and has no significant effect on the level of technical inefficiency in lowland rice farming. The results of this study are in line with research conducted by Saptana (2011) that the ratio of the number of working-age household members to the total household members has a negative but not significant effect on the technical inefficiency of curly red chili farming in Central Java Province.

The variable distance between the land and the farmer's house (Z5) is positive and has a significant effect on the technical inefficiency of lowland rice farming. Farmers who are close can control farming at any time, while farmers who have more land to farm will find it more difficult to control their farming so the level of technical efficiency of farming can be reduced. Muslimin's research (2012) found that the distance between the farm and the farmer's house has a negative and significant effect on increasing the technical inefficiency of farming.

Production risk control model

The optimal lowland rice farming production function model can be obtained by optimizing the productivity function, minimizing the production risk function, and the source of technical inefficiency with the following model:

$$TP = f(E_T) - F(\sigma^2) - F(Z)$$

$$f(E_T) = \mathbf{0{,}1258 + 0{,}1935\ lnBE + 0{,}3742\ lnPN + 0{,}1615\ lnPP + 0{,}0546\ lnPK + 0{,}1093\ lnPO + 0{,}1267\ lnPT + 0{,}1856\ lnPJA}$$

$$F(\sigma^2) = \mathbf{0{,}1258 - 0{,}3654\ lnBE^* - 0{,}4362\ lnPN^* - 0{,}1534\ lnPP^* - 0{,}0562\ lnPK^* - 0{,}0815\ lnPO^* - 0{,}1252\ lnPT^* + 0{,}0765\ lnPJA^*}$$

$$F(Z) = \mathbf{0{,}4876 + 0{,}1487\ lnLT^{**} - 0{,}1514\ lnBE^{**} - 0{,}0451\ lnPN^{**} - 0{,}1145\ lnPP^{**} - 0{,}0676\ lnPK^{**}}$$

IV. CONCLUSION

Cultivation technology carried out by farmers is still conventional with the use of production inputs that are still below the recommended dose. The productivity obtained by farmers is low, and the production risk is high. The determinants of the productivity function of lowland rice farming are urea, SP36, KCL, and organic fertilizers. Optimal use of these production inputs will be able to reduce the occurrence of production risk. Farmers' behavior in responding to production risks is to avoid risk and is mainly determined by the demand for urea and SP36 fertilizers. Sources of technical inefficiency mainly come from a narrow land area. Production risk control can be done by increasing productivity through the use of optimal production inputs, especially urea, SP36, and organic fertilizers, as well as an intensification of land area.

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Effect of elevated carbon dioxide on growth and development of *Santalum album* L. seedlings inoculated with plant growth promoting microorganisms in Open Top Chambers

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***Abstract*— *Rising carbon dioxide (CO_2) concentrations in atmosphere have a significant impact on plant growth and metabolism. Many plant growth promoting microbes play an important role in maintaining plant growth and vitality by facilitating nutrient translocation under adverse conditions. This study was carried out to investigate the effects of elevated CO_2 on growth parameters (height of the seedlings, increment in height, collar diameter and number of leaves) in sandalwood (Santalum album L.) seedlings inoculated with beneficial microorganisms (*Pseudomonas *putida*, Bacillus *subtilis* and Trichoderma *harzianum) in the Open Top Chambers (OTCs). Sandalwood seedlings inoculated with* P. *putida*, B. *subtilis*, T. *harzianum were grown in OTCs at 600 ppm, 800ppm, 1000ppm and 1200ppm elevated concentrations of CO_2. As compared to the non-inoculated (untreated/control) plants, inoculated one shows a high growth and development rate but non- inoculated plants were not able to handle stress at higher/elevated CO_2 concentration (1000 & 1200ppm) and ultimately died after 60 days. P. putida showed a high growth rate on all growth and development parameters taken up to 1000 ppm concentration of CO_2 followed by Bacillus subtilis. It is also observed that T. harzianum treatment could not withstand the elevated concentration of CO_2 beyond 1000 to 1200 ppm whereas P. putida treatment was found to be effective at even at 1200 ppm of CO_2. Through these experiments under OTCs at different elevated concentrations of CO_2, we can predict the possibility of climate change and global warming effect on beneficial microbes and vis-a-vis their effect on growth, development and yield on crops. Hence, other plant growth microbes including mycorrhizal fungi can be analyzed for future research and bio-prospect under OTCs experimentation.*

Keywords— ***elevated carbon dioxide; open top chambers; plant growth microbes; sandalwood.***

I. INTRODUCTION

Santalum album or tropical Indian sandalwood and it is one of the members of genus, *Santalum*, comprises of 16 species which are economically important and distributed globally (Shea *et al.* 1998). *S. album* is immensely famous for its essential oil having santalol content but due to lack of sizeable trees, it is no longer used for fine woodworking as before (Srinivasan *et al.* 1992; Radomiljac and McComb 1999). The wood is commercially known as "East Indian Sandalwood" and internationally reflects as

“Dollar earning parasite" (Durairaj and Kamaraj 2013). It has a history of more than 5000 years and India has been a hub for production and use of sandalwood in its different forms particularly sandalwood oil for medicine, cosmetics and perfumery (Hansda 2009).

Santalum album is a partial root parasite which requires a host for nutritional fulfillment. The growth is severely affected in absence of a proper host. The *Santalum* roots produce a specialized structure called ‘haustoria’ for deriving the nutrients such as nitrogen and phosphorous from the soil (Teixeira da Silva *et al.* 2016), although, it is able to absorb calcium and potassium in a good manner (Iyengar 1965). Anthropogenic activities and emissions enhance the atmospheric carbon dioxide level which would result in global warming (Beardall and Raven 2004). Increased carbon dioxide (CO_2) level also have an impact on *S. album* growth parameters such as collar diameter, seedling height, number of leaves, photosynthetic rate and transpiration rate The rising CO_2 concentration has a direct and indirect link with the growth and metabolism of plants and rhizospheric microbes as CO_2 is a primary raw material in the process of photosynthesis. Many strains of bacteria and fungi enhance plant growth through bio-inoculation, a method of introducing a microbial culture into the rhizosphere of the plants to maintain vitality and healthy growth. Hence, Plant Growth Promoting Rhizobacteria (PGPR) hold a special place whenever there is a talk of ‘bio-fertilizers’ in general and particular.. But due to the increasing amount of carbon dioxide in atmosphere, there is a need to study the effect caused in the microbiota/microflora due to this factor. Open Top Chambers (OTCs) offers micro environment surroundings, lower light intensity, higher relative humidity and a constant wind velocity (Leadley and Drake 1993). Hence, OTCs can be used to detect carbon dioxide and its effect on beneficial microbes and plants to predict the climatic change effect going to happen in near future. Therefore, a study was carried out to see the effect of elevated carbon dioxide on growth and development of *S. album* seedlings inoculated with plant growth promoting microorganisms in OTCs.

II. MATERIAL AND METHODS

2.1. Study area:

The area for study was ‘New Forest Campus’, Forest Research Institute (FRI), Dehradun, Uttarakhand, India. It is situated in Doon valley and this area is surrounded by west Himalayan ranges in north and Shiwalik ranges in south running parallel to it. This campus covers an area of 4.45 km^2. It lies at an elevation of 660 m above sea level. The annual rainfall is over 200 cm, bulk of which falls as south-west monsoon from June to September. The temperature ranges between 1 to 42° Celsius in winters to summers. The annual mean temperature stabilizes to 20 degree Celsius.

2.2. Collection of plants:

The sandalwood plants/seedlings were grown and collected from the Central Nursery, FRI, Dehradun. The seedlings were checked for any infection which was already either present in root trainers or plastic bags. The soil was also collected from the Central Nursery and used in sterilized condition in the plantation of seedlings in the OTCs.

2.3. Culturing of microorganisms for bio-inoculation:

Two bacterial strains *i.e. Pseudomonas putida* (Pp-1), *Bacillus subtilis* (Bs-I) and one fungal strain/species *i.e. Trichoderma harzianum* (Th-I) were taken to study the response of plants/seedlings inoculated with them as these microbes are found in rhizosphere and help in nutrient mobilization in the rhizpspheric region (Figure 4)

Serial dilution method was adopted for isolation of bacteria (Johnson and Curl 1972) and fungi (Waksman 1927) from soil samples collected from the rhizosphere. The fungal and bacterial species were identified based on several biochemical tests and their morphological features (Bergey *et al.* 1934; Rifai 1969; ABIS online- Bacterial identification - ABIS online respectively.

2.4. Mass multiplication of microbial cultures:

2.4.1. *Pseudomonas putida* and *Bacillus subtilis*

The culture was mass multiplied in Nutrient Broth (NB) medium (Figure 5). The designated amount of nutrient broth was dissolved fully in 300 ml of distilled water. The nutrient media was then autoclaved under high pressure for 20-25 minutes. This process must ensure that the media is free of any contamination. A master culture was prepared by inoculating them under the Laminar Air Flow (LAF) chamber and incubating the culture for a couple of days. Also for checking the maximum growth period, 9 ml of autoclaved NB medium was taken in test tubes. These were inoculated with 1ml of target bacterial culture. The test tubes were then kept in B.O.D. shaker *cum* incubator. Each test tube was then monitored every hour with the growth of the inoculation. For this process, absorbance of each test tube solution was taken every hour through a UV – VIS Spectrophotometer at 600 nm. Results indicated that maximum growth for *Bacillus subtilis* was found at 10:30 hours and that of *Pseudomonas putida* at 11:30 hours of duration. This step also helped to know the incubation period for mass culture of these bacteria for further experiments.

2.4.2. *Trichoderma harzianum* multiplication

A mixture of saw dust, wheat bran and sterilized distilled water was made in the ratio of 1: 3: 4. The circular pieces of inoculum so isolated were then placed into each of the bag of mixture. The culture was then put in a B.O.D. (Biological Oxygen Demand) incubator at a temperature of 27° C ± 2° C for three to four days to obtain the mass culture for *Trichoderma harzianum*. The CFUs (Colony Forming Units) analysis for the saw dust mixture revealed that the mixture has 3.4×10^7 colony forming units per gram in it (Figures 5 & 6).

2.5. Inoculation of microbes into the rhizosphere of Sandalwood seedlings:

Three healthy seedlings of same height were planted safely along with the host (*Alternanthera* species) in poly bags (12 × 12 inch). The soil so used was first autoclaved. For bacterial inoculations, which are in liquid state, the inoculation so used was ten percent of the soil used. For each seedling 50 ml of inoculum was used in 500 g soil. For *Trichoderma* inoculation the inoculum was weighed to ten percent of soil. The sawdust mixture was mixed with the soil in each poly bag to help in the inoculation of the fungus in the rhizosphere of the plants.

2.6 Experimental design and statistical analysis:

The OTCs were already been adjusted to provide a desired concentration of carbon dioxide in the chamber (size, L×W×H; 3×3×4 m with 40-50% humidity and 27-29 ºC temperature) which can be adjusted as well and can alters the microenvironment of the plants. The optimum concentration of CO_2 in Doon valley was found to be 440 ppm (Dehradun, Uttarakhand, India). These chambers were provided/fixed with elevated 600, 800, 1000, 1200 ppm concentrations, hence, seedlings may respond to the altered microclimate in these chambers through various mechanisms like morphological, anatomical or physiological changes. The growth parameters taken under consideration were increment in height, collar diameter, number of leaves and microbial rhizospheric colonization/population in $CFUml^{-1}$. The experimental design opted for the purpose was CRD (Completely Randomized Design) with three replicates. The data were statistically analyzed.

III. RESULTS AND DISCUSSION

Sandalwood seedlings were kept in the carbon dioxide rich atmosphere *i.e.* OTCs and different growth parameters were studied under various inoculated treatments till 90 days.

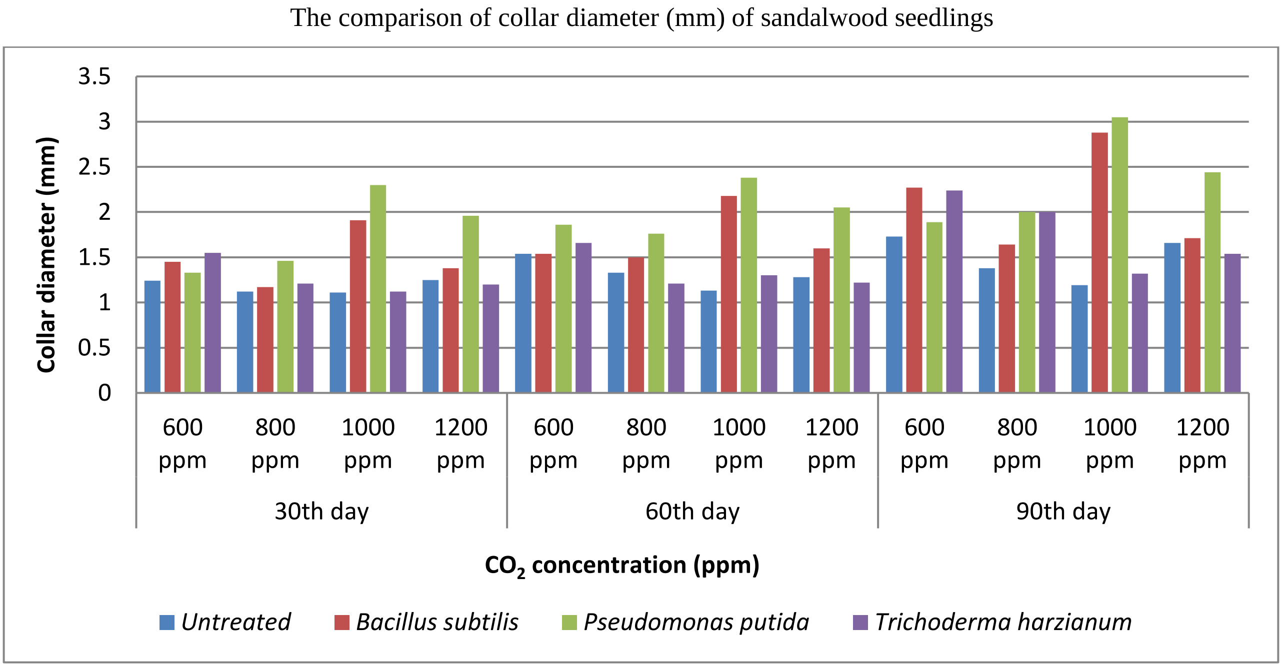

Fig.1. A histogram graphical representation of comparison of collar diameter (mm) of sandalwood seedlings after the 30th, 60th and 90th day under different treatments.

After 30 days of inoculation, the *Pseudomonas putida* treatment showed prominent increment in collar diameter in comparison to all other treatments including the untreated/control treatment. *Trichoderma harizianum* treatment showed greater collar diameter at CO_2 concentration of 600 ppm. On the other hand, the collar diameter in *P. putida* treatment was maximum at 800, 1000, and 1200 ppm of CO_2 concentrations than other treatments after 30 days (Fig. 1).

The most important finding after 60 days of observations was that the collar diameter in *Pseudomonas putida* treatment was greater at different concentrations *i.e.* 600 ppm, 800 ppm, 1000 ppm, and 1200 ppm of CO_2 than other treatments. In this finding, except for the 600ppm concentration of CO_2, all the concentrations of CO_2 in *P. putida* treatment showed the increased collar diameter as compared to other treatments; however, the untreated/control seedlings have lesser collar diameter as compared to other treatments. The same finding was found after 90 days of observation (Fig. 1).

After 90 days of inoculation, there has been a significant increase in collar diameter of the seedlings with inoculation than control (untreated) seedlings. The maximum enhancement in collar diameter growth is shown by the sandalwood seedlings inoculated with *Pseudomonas putida* at 1000 ppm of CO_2 concentration. The seedlings inoculated with *Bacillus subtilis* also showed a good increasing trend in collar diameter measurement. Out of the all treatments, *Pseudomonas putida* also showed a good expansion in diameter at 1200 ppm of CO_2 concentration which is not seen so pronounced in the other treatments. The untreated (control) seedlings have the minimum collar diameter (1.8-2.2 mm) than other treatments (Fig. 1).

The comparison of number of leaves in sandalwood seedlings

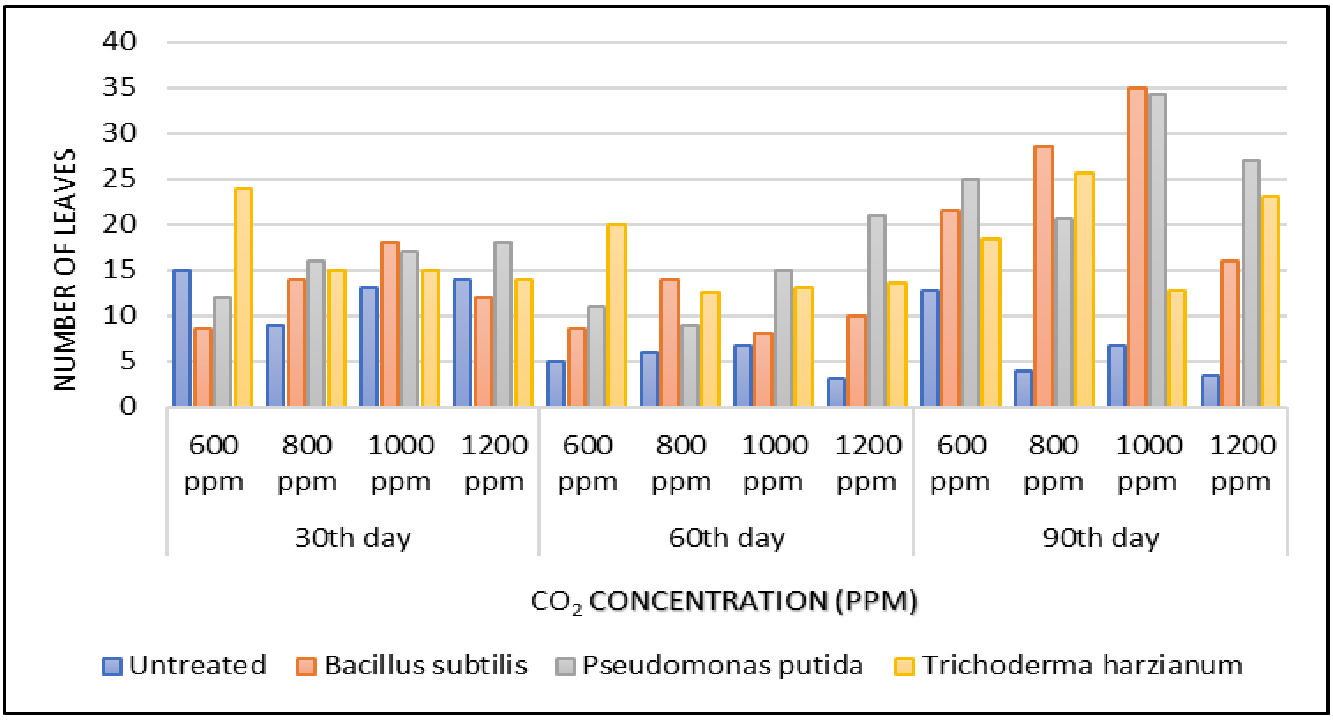

Fig.2. A histogram graphical representation of comparison of number of leaves of sandalwood seedlings after 30th, 60th and 90th day under different treatments.

Although, *Trichoderma harzianum* treatment showed more number of leaves per plant at 600 ppm CO_2 concentration after 30 days of inoculation but *Bacillus subtilis* and *Pseudomonas putida* treatments produced lower numbers of leaves per plant than the control seedlings at 600 ppm CO_2 concentration after 30 days of inoculation. At 1200 ppm CO_2 concentrations, *Pseudomonas putida* treatment showed more number of leaves per plant than other treatments.

After 60 days of inoculation, *Trichoderma harzianum* treatment had more number of leaves, whereas *Pseudomonas putida* treatment had maximum number of leaves at 1200 ppm, concentrations than other treatments. The control seedlings have less number of leaves than other treatments (Fig.2).

Bacillus subtilis and *Pseudomonas putida* treatments exhibited the same number of leaves at 1000 ppm CO_2 concentration after 90 days of observation. Whatsoever, these findings have revealed that when we compared the data with the control or other treatments then *P. putida* treatment seedlings were growing at their fastest rate at 1000 ppm and 1200 ppm of CO_2 concentrations. The inoculated seedlings showed an enhanced resilience to stresses which is exhibited through an increase in growth parameters. Each type of microorganisms has its own 'niche' of performance. *Trichoderma harzianum* treatment performs best at 800 ppm and those of both bacterial strains in treatments perform best at 600ppm and 1000 ppm of CO_2 concentration respectively (Fig. 2).

The comparison of increment in height of sandalwood seedlings

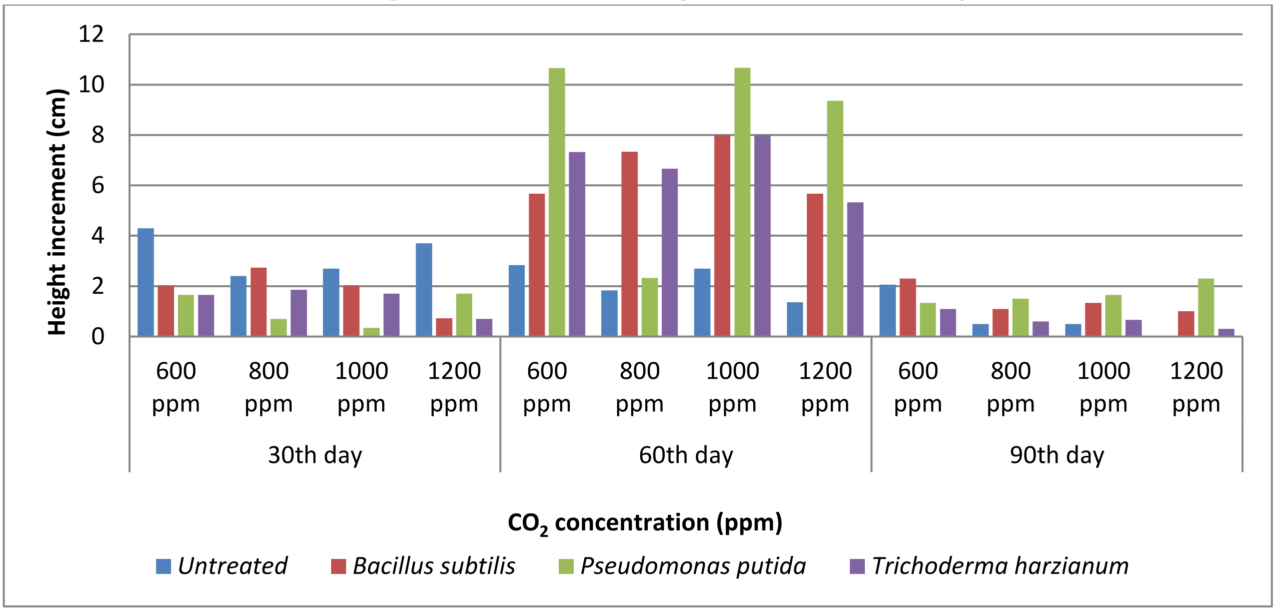

Fig.3. A histogram graphical representation of comparison of increase in height of sandalwood seedlings after 30th, 60th and 90th day under different treatments. The data was analysed statistically for analysis of variance. There was a significant growth effect of all bioinoculants on seedlings at P <0.05 for different time intervals (F=4.94; p=0.027).

After 30 days of observation, the increment in seedling height at 600 ppm, 1000 ppm, and 1200 ppm of CO_2 concentrations was maximum in the control/untreated treatment as compared to the other treatments, but at 800 ppm CO_2 concentration, the *Bacillus subtilis* treatment exceeded the control in height increment. After 60 and 90-days of inoculation period, the *Pseudomonas putida* treatment showed increment in height of seedlings as compared to control treatment (Fig. 3). Increment in height of sandalwood seedlings inoculated with *Pseudomonas putida* showed preeminent results in comparison to other treatments as far as increment in height is concerned followed by *Bacillus subtilis* treatment.

Bacillus subtilis also showed good effect on height in comparison to control/untreated treatment. Inoculation with *Trichoderma harzianum* treatment also exihibited height increment than the untreated sandalwood seedlings but pace of growth was rather slow as compared to *Pseudomonas putida* treatment (Figure 6).

Effect on microbial bioagents population due to high CO_2 concentration in OTCs after 45 days, the CFUml^{-1} count varies at different concentrations of CO_2. At different concentrations of CO_2 (*i.e.* 600ppm, 800ppm, 1000ppm, and 1200ppm), the highest CFUml^{-1} count was seen in *Pseudomonas putida* treatment than *Bacillus subtillis* treatment where the CFU.ml^{-1}count was higher than in *Trichoderma harzianum* treatment.

Table 1. Effect on microbial bioagents population due to elevated CO_2 concentration in OTCs after 45 Days

Duration	CO_2 Concentration (ppm) in OTCs	Control (CFUml^{-1})	*Bacillus subtilis* (CFUml^{-1})	*Pseudomonas putida* (CFUml^{-1})	*Trichoderma harzianum* (CFUml^{-1})
45 Days	600	0	67 ×10^3	76 × 10^3	27 × 10^3
	800	0	60 × 10^3	72 × 10^3	18 × 10^3
	1000	0	33 × 10^3	70 × 10^3	9 × 10^3
	1200	0	22 × 10^3	68 × 10^3	7 × 10^3

Table 2. Effect on Microbial bioagents population due to elevated CO_2 concentration in OTCs after 90 Days

Duration	CO_2 Concentration (ppm) in OTCs	Control ($CFUml^{-1}$)	*Bacillus subtilis* ($CFUml^{-1}$)	*Pseudomonas putida* ($CFUml^{-1}$)	*Trichoderma harzianum* ($CFUml^{-1}$)
90 Days	600	0	69×10^3	77×10^3	25×10^3
	800	0	63×10^3	70×10^3	16×10^3
	1000	0	30×10^3	70×10^3	6×10^3
	1200	0	10×10^3	61×10^3	3×10^3

OTC - Open Top Chambers, CFU - Colony Forming Units

Although, after 90 days of inoculation, the number of bacterial colonies have grown slightly but this is due to the fact that the bacteria exhibited stationary phase which indicates that they haven't divided but are still metabolically active in the rhizosphere of inoculated seedlings. The houstoria were also seen in the roots association of both host and *Santalum album* seedlings (Figure 5). The control/untreated seedlings do not have any bacterial and fungal colony as no bioagents was put into the control set in sterilized soil condition (Tables 1&2).

Effect on microbial bioagents population due to elevated CO_2 concentration in OTCs after 45 and 90 days are shown in following Tables 1 & 2.

Plants exposed to elevated CO_2 often show increased growth and water use efficiency (Rogers and Dahlman 1993; Allen and Amthor 1995; Wittwer 1995) and increased rates of photosynthesis (Long and Drake 1992; Amthor 1995). Plants exposed to elevated atmospheric CO_2 are almost always larger than those grown in ambient CO_2. The magnitude of growth stimulation is typically dependent upon photosynthetic pathway, sink strength, phenotypic plasticity and plant life history strategies (Hunt *et al.* 1991). Elevated CO_2 also increased the photosynthesis rates of young and fully expanded leaves by 35—46% and of whole plants by more than 50% (Ryle *et al.*, 1992). The most surprising feature of the experimental results was that the observed increases in rates of leaf and whole plant photosynthesis in elevated CO_2 had, relatively, a very small effect on plant growth (Ryle *et al.* 1992). Elevated CO_2 affected plant weight in the first 10-20 days but development constrained the branch numbers. In this study also, when we compared the data with the control or other treatments then *P. putida* treatment seedlings were growing at the fastest rate at higher 1000 ppm and 1200 ppm of CO_2 concentrations. Subsequently, according to Ryle *et al.* (1992) when mature leaf axils provide a potential increase in tillers, it might be expected that plant weight would have accelerated even faster in elevated CO_2. This never occurred, for tiller numbers from both CO_2 concentrations increased. This imposed a severe restriction on potential growth in elevated CO_2 because the weight increases which can be achieved on a single axis are constrained by the environmental and ontogenetic control of leaf length and width and ability to store unused carbohydrate.

Growth analysis of two *Eucalyptus* species (*e.g. Eucalyptus macrorhyncha* and *Eucalyptus rossii*) indicated that increased CO_2 may allow *Eucalyptus* species to perform better during conditions of low soil moisture. Tissue *et al.* (1993) observed down-regulation of photosynthetic capacity in seedlings grown in elevated CO_2 when well-watered but not when water stressed. The down-regulation of photosynthesis of plants grown in elevated CO_2 is often associated with starch accumulation (Tissue *et al.*, 1993). Plants grown in elevated CO_2 had greater leaf, stem and total biomass than plants grown in ambient CO_2. Similar findings are observed in this experiment where *Trichoderma harzianum* treated seedlings had more number of leaves in general, whereas *Pseudomonas putida* treated seedlings had maximum number of leaves at 1200 ppm, concentrations respectively. The control seedlings have less number of leaves than other treatments.

Although, there is a report on elevated CO_2 that it had had no effect on the vegetative attributes of *Cardamine hirsuta*, *Spergula arvensis* and *Poa annua* whereas *Senecio vulgaris* produced longer leaves and greater biomass. Both *Senecio* and *Poa* species had faster maturation times. The vegetative response of *Senecio vulgaris* was not translated into increased seed output, although, seed mass and carbon: nitrogen ratios were significantly increased. By contrast, *Poa* species showed no vegetative response to elevated CO_2, but had significantly increased seed production. Maximum biomass may be achieved under elevated CO_2 when other resources are not limiting, the relative enhancement of biomass owing to elevated CO_2 may be greatest under conditions of low resources, such as light (Zangerl and

Bazzaz 1984; Bazzaz and Miao 1993). None of the above four plant species showed responses to elevated CO_2 during germination and early growth (up to 16 days after sowing).There were no differences in the early growth (16 days post-sowing) responses of the four-plant species to elevated CO_2. Carbon dioxide concentrations near the soil surface may not be well correlated with canopy or atmospheric concentrations. The CO_2 concentrations near the soil surface being far higher because of soil microbial activity than at greater heights in the canopy (Bazzaz and Williams 1991) and hence, elevated CO_2 significantly increased the total biomass of the seedlings. The combined effect of the elevated CO_2 and temperature treatments further increased the total biomass, but not significantly. There are reports that the content of nitrogen and water decreased, while some secondary compounds (such as condensed tannins and flavanols glycosides) increased in leaves subjected to CO_2 enrichment (Kuokkanen *et al.* 2001).

The total biomass of the seedlings was increased by CO_2 enrichment but not by the temperature (Kellomäki and Wang 1998). The seedling biomass in the field control was significantly smaller than that in the control chambers. The nitrogen and water content were significantly decreased by CO_2 enrichment, but not by increased temperature or an increase in both factors. The greatest amount of biomass was produced in the elevated CO_2 and elevated temperature combination. However, this result supports the widely held prediction that the growth of forest trees in the boreal zone will be enhanced by the "fertilization effect" of CO_2 and lengthening of the growth season as result of higher air temperatures (2–4° C) (Kellomäki and Väisänen 1997; Kellomäki and Wang 1998). A doubling of the atmospheric CO_2 concentration enhanced plant growth and significantly increased stomatal index also. However, there was no significant change in relative stomatal density in *Alnus glutinosa* plants grown in the elevated CO_2 concentration showed an overall general increase in growth of the measured parameters relative to those grown at ambient CO_2. There was a significant increase in the number of branches and a decrease in specific leaf area with elevated CO_2, but no significant increase in plant height, number of leaves and absolute leaf area. This supports the general observation that an increase in atmospheric CO_2 concentration enhances overall plant growth (McKee *et al.* 1995; Long *et al.* 1996; Mulholland *et al.* 1998). In this study also, it is found that the higher concentration of CO_2 in control chambers suppressed the plant height and development than inoculated one but only *Psedomanas putida* and *Bacillus subtilis* treatments withstand the higher concentration of CO_2 and resulted in the good growth of inoculated seedlings. Whereas, the *Trichoderma harzianum* inoculated seedlings had lower growth effect due to elevated CO_2 level (1000-1200ppm) concentration. It is also evident from the microbial population study from the rhizosphere of inoculated seedlings after 90 days of inoculation that the bacteria especially *P. putida* is more resistant to elevated CO_2 concentration followed by *B. subtilis*. Whereas, the fungal bioagent *i.e. T. harzianum* is vulnerable at higher concentration of elevated CO_2 (1000-1200 ppm) than bacterial bioagents *i.e. P. putida* and *B. subtilis*. It might be due to endospore formation in bacterial bioagents than fungal bioagent as a result of elevated CO_2 concentration stress.

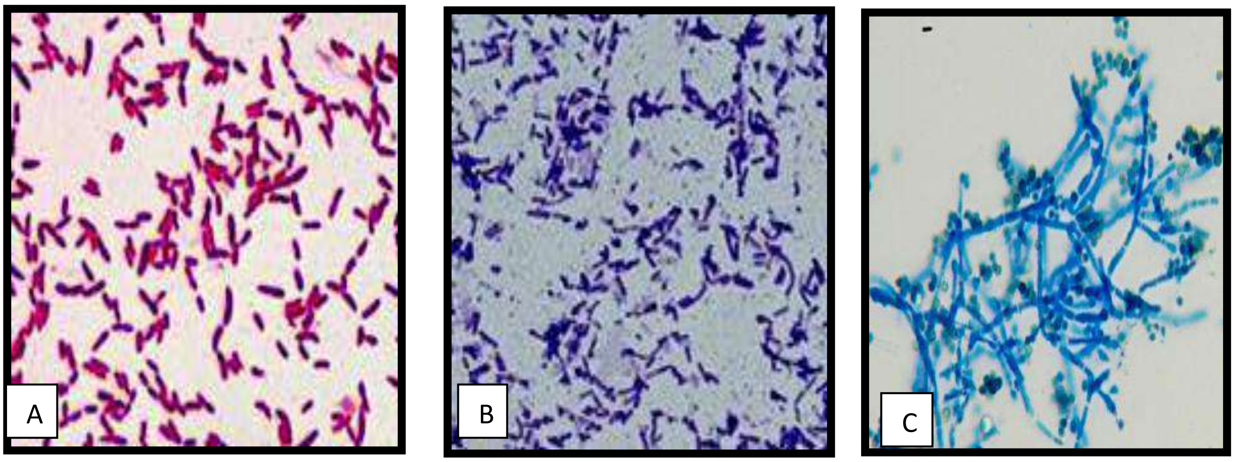

Fig.4. Micrographs of microbial inoculants; A. Pseudomonas putida Gram negative, rod shaped bacteria; B. Bacillus subtilis, Gram positive, rod shaped bacteria; C. Trichoderma harzianum fungus with conidia

Fig.5. A. One year Sandalwood plant B. Inflorescence; C. Sandalwood fruits; D. Root-Root association of Sandalwood and host plant; Sandal plant (yellow arrow) and the pot host plant (Alternanthera species; red arrow); E. Haustoria as seen in root trainer conditions; F. Isolated haustoria showing both host and parasite roots; G. Haustoria (zoomed) H. Bacterial cultures I. Trichoderma harzianum culture J. Mass culture of Pseudomonas putida and Bacillus subtilis

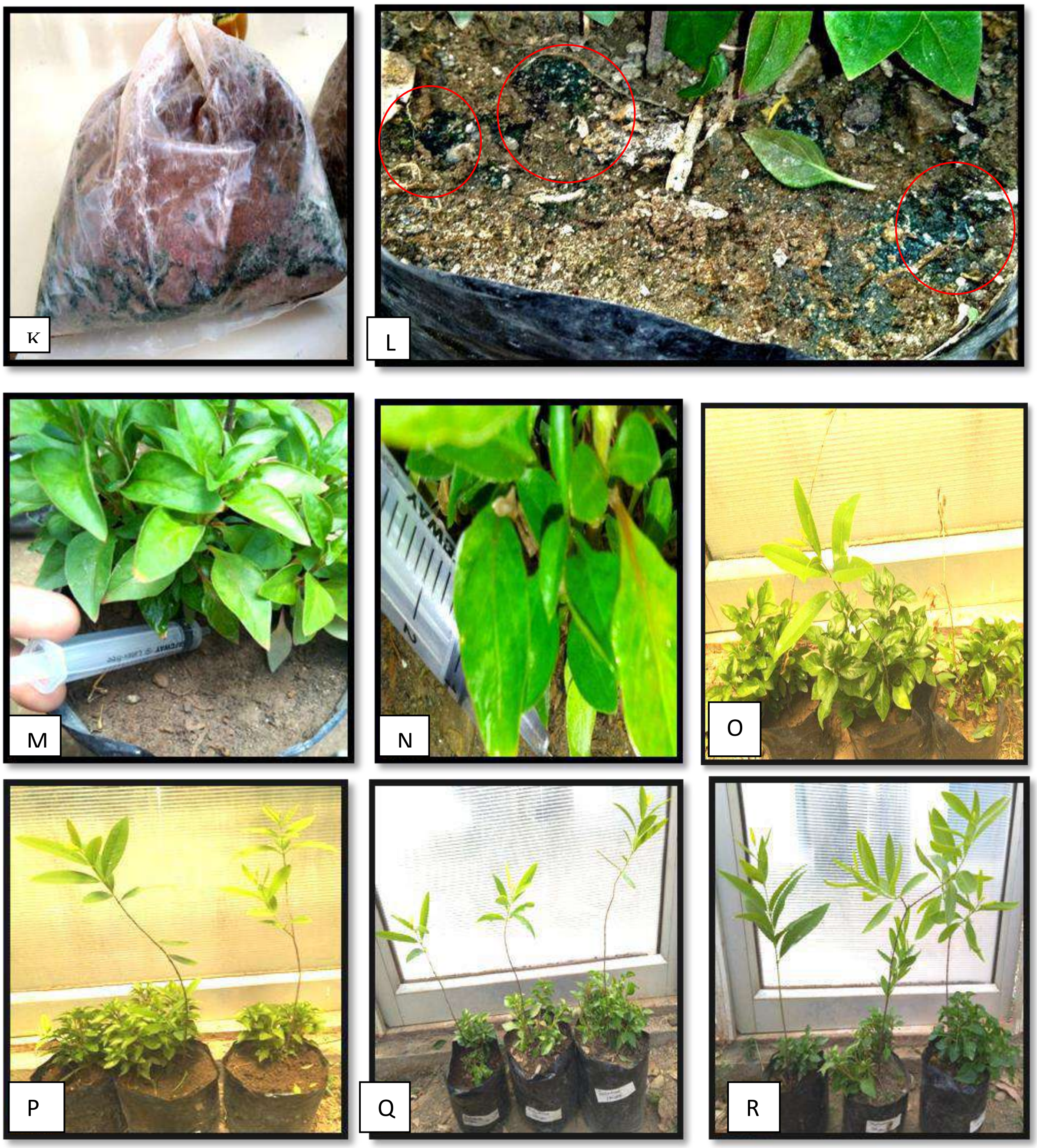

Fig.6. K. Trichoderma harzianum culture in saw dust mixture; L. Green patches showing Trichoderma harzianum (in red circles) inoculation; M-N. Putting bacterial cultures/inoculants in the Sandalwood rhizosphere; O-R. Sandalwood seedlings under treatment in different OTCs (O- Control; P- Trichoderma harzianum; Q- Bacillus subtilis; R- Pseudomonas putida)

IV. CONCLUSION

In the present study, *Pseudomonas putida* treatment showed a high growth rate on growth parameters up to the increased concentration of 1000 ppm of CO_2 in comparison to another treatments and untreated control. But further increment in CO_2 concentration up to 1200 ppm, the growth of seedlings is enhanced, but the pace of growth slowed down a little bit. The seedlings without any inoculation showed an increment in growth till 30 days only after which stagnation in growth of the seedlings has been seen. The plants were not able to handle stress at higher CO_2 concentration and ultimately they die after 60 -days in control/untreated set. But the seedlings inoculated with microbial inoculants withstood the stress of higher concentration of CO_2 and showed good growth and development. The study can also be used to predict the effect of increasing carbon dioxide concentration in the atmosphere on the growth and development of plants. This can also help to know the saturation effect of carbon dioxide on vegetation also. Although, it is indicating that

'CO_2 chamber fertilization effect' does not always hold true. In the wake of climate change phenomenon, there is a talk of adaptive and mitigation strategies to enhance the resilience of a plant species to reduce its vulnerability. 'Bio-fertilization' can help us on this front by serving the dual purpose of enhancing the growth as well as resilience of a plant species. Nevertheless, this type of study has been less conducted in forestry plant species/crops and it is only a small step taken in the direction to measure the effect of microbial bio- fertilization on this aspect.

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The Relationship of Bivalvia Affairs to Mangrove Density in Pt Arutmin Indonesia Kintap Mining Area Mekarsari Village, Tanah Laut Regency South Kalimantan Province

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***Abstract*— *This research was conducted to determine how strong the effect of mangrove density on the bivalves community in particular in abundance. This research was conducted in March 2022 to April 2022. Data collection is carried out at three stations located at upstream, middle and downstream. Each sample station collected is a bivalves sample and data mangrove observation. Bivalve samples using a 1 mx 1 m transect on the plot observation of mangroves 10 m x 10 m. The results of the study showed that the bivalves found there are 8 types of bivalves, namely; Mactra violacea, Perna viridis, Mactra chinensis, Polymesoda erosa, Geloina expansa, Isognomon ephippium, Anadara antiquata and Polymesoda bengalensis. Station 1 with bivalves abundance of 960 ind/m² with mangrove density 1300 ind/ha. Station 2 with an abundance of 680 ind/m² bivalves with a mangrove density 1800 ind/ha. Stations with an abundance of bivalves of 2,360 ind/m² with a density of mangroves 2200 ind/ha. The relationship between bivalves abundance and mangrove density using simple linear regression analysis with weak category results*

***Keywords*— Bivalves, Mangrove, Mekarsari Village**

I. INTRODUCTION

Mangrove ecosystems along the coast or river estuaries are affected by tides and live in coastal areas that are protected from the waves. Likewise, the mangrove ecosystem in Mekarsari Village, which is located in the Kintap sub-district, Tanah Laut district, South Kalimantan Province is an area that has a 1.83 ha mangrove ecosystem (Tony. F, et al 2022). The ecological function of the mangrove ecosystem is as a beach protector from wind, waves and currents from the sea, habitat, foraging areas, nursery areas and spawning areas (Samir and Romy, 2016).

One of the communities found in the mangrove ecosystem is the bivalves community. The bivalve community occupies an important position in the food chain in the mangrove forest area because bivalves are detritus-eating organisms and have a role in the decomposition process of mangrove litter. In addition, it can also be used as a pollutant indicator because it has a long life cycle, can be identified and its abundance can be calculated (Rosenberg and Resh, 1993).

Research (Defira, 2018) states that mangrove density affects the abundance of individual bivalves. The higher the density of mangroves, the higher the abundance of bivalves because they are able to produce high litter and organic matter needed by bivalves for food sources. So this study was conducted to determine how strong the effect of mangrove density on bivalves communities, especially on their abundance.

Based on the problem formulation, how is the relationship between the abundance of bivalves and the density of mangroves in Mekarsari Village. The purpose of this study was to analyze the relationship between the abundance of bivalves and the density of mangroves in Mekarsari

Village. This research can be used as a reference for future research.

II. RESEARCH METHODS

This research was conducted in March – April 2022 in the PT Arutmin Indonesia Tambang Kintap area, Mekarsari Village, Tanah Laut Regency, South Kalimantan Province. The equipment used during the study were GPS, stationery, camera, bivalve and mangrove identification book, plastic bag, tissue, label paper, meter, pipette, tally sheet, 1 m × 1 m quadrant transect, shovel, arcgis 10.8 and microsoft excel. While the materials used are distilled water, 10% formalin and bivalves samples.

Determination of observation points using purposive sampling method. Purposive sampling is a sampling method based on research considerations to determine the characteristics of a population that has a dominant relationship so that research objectives can be achieved. There are 3 designated observation points, namely station 1 which is located directly opposite the sea or commonly known as open mangrove. Stations 2 and 3 are brackish mangrove areas along the river.

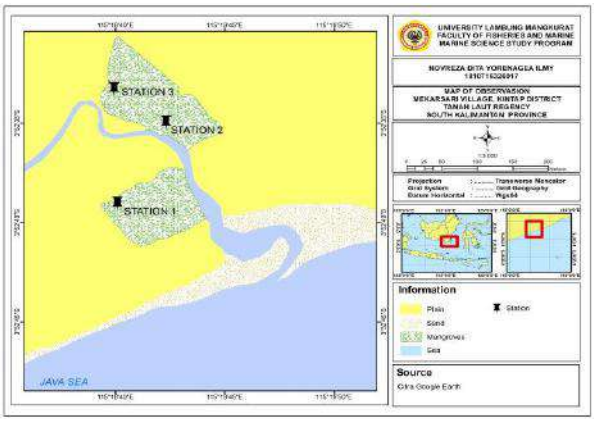

Fig.2.1. Observation Point

Bivalves sampling was carried out on each 10 m x 10 m mangrove observation point plot. Bivalve sampling was taken at five points to represent the plot area of 10 m x 10 m, four points at the corners of each plot and one point in the center of the plot using a 1 m x 1 m transect. Bivalves are taken by hand for those on the surface and using a shovel for bivalves that are in the substrate by digging 10-15 cm.

Sampling of mangroves determines the mangrove observation path starting with a perpendicular direction from the sea to the land along the existence of mangroves. Transect installation was carried out at three stations with a distance of 50 meters between transects. Make a transect using a 10 m × 10 m rapia rope and observe the mangroves with tree criteria, namely diameter at chest height 10 cm. Mangrove samples found in each observation plot, counted the number of stands of each species.

Bivalve Data Analysis

The number of individuals of a species to the total number of individuals contained in an area in a community, is formulated as follows:

$$K = \frac{ni}{5} \times 100$$

Information:
K = Species Abundance (ind/m2)
ni = Number of individual bivalves (ind)
5 = Number of Sample Plots
100 = Total Sampling Area (m2)

The relative abundance of bivalves was calculated using the Shannon Wienner formula (Odum, 1993), with the following formula:

$$R = \frac{ni}{N} \times 100\%$$

Information:

R = Relative Abundance (ind)

ni = Number of individuals of each species (ind)

N = Total Number of Individuals (ind)

Analisis Data Mangrove

Species density is that in a unit area there are the number of stands of species i. standing category based on growth category, namely trees with diameter criteria at chest height > 10 cm. The formula for specific density is as follows:

$$K = \frac{ni}{A}$$

Information:

K = Specific Density I (ind/m2)

ni = Total Stands of Type I (eng)

A = Total Sampling Area (m2)

The relative density of species is the ratio of the total number of stands of species i to the total number of all stands of species, the following formula is used:

$$KRi = \frac{ni}{\sum n} \times 100\%$$

Information:
KRi = Specific Relative Density (%)
ni = Total Number of Stands of Type I (ind)
$\sum n$ = Total Number of Stands of all Types (ind)

III. RESULTS AND DISCUSSION

Based on the results of research that has been carried out, it is known that 8 species of bivalves found in the mangrove area of Mekarsari Village include; Mactra violacea, Perna viridis, Mactra chinensis, Polymesoda erosa, Geloina expansa, Isognomon ephippium, Anadara antiquata and Polymesoda bengalensis. The composition of bivalves can be seen per station in Table 3.1.

Table 3.1. Bivalve Type Composition

Species Name	Station		
	1	2	3
Mactra violacea	+	+	+
Perna viridis	+	-	-
Mactra chinensis	+	+	-
Polymesoda erosa	-	-	+
Geloina expansa	+	+	+
Isognomon ephippium	+	-	-
Anadara antiquata	-	-	+
Polymesoda bengalensis	+	+	+

(*Source: Primary Data 2022*)

Informationn :

\+ : Found

\- : Not Found

The distribution of bivalves varies or varies at each observation station in general, this is due to differences in habitat characteristics for each type of bivalves, availability of food sources and tolerance to environmental changes..

Abundance

Based on the results of research that has been carried out the abundance of bivalves found at station one can be seen in Table 3.2.

Table 3.2. Data Result of Analysis of Abundance of Bivalves Station 1

Station	Species	(ind)	K (ind/m^2)	KR (ind)
	Perna viridis	3	60	16,67
	Mactra chinensis	7	140	38,89
1 1	*Isognomon ephippium*	4	80	22,22
	Polymesoda bengalensis	4	80	22,22
	Mactra violacea	3	60	21,43
	Perna viridis	1	20	7,14
	Mactra chinensis	3	60	21,43
2	*Geloina expansa*	3	60	21,43
	Isognomon ephippium	1	20	7,14
	Polymesoda bengalensis	3	60	21,43
	Mactra violacea	6	120	37,5
	Perna viridis	2	40	12,5
	Mactra chinensis	1	20	6,25
3	*Geloina expansa*	2	40	12,5
	Isognomon ephippium	1	20	6,25
	Polymesoda bengalensis	4	80	25
Total		48	960	300

(*Source: Primary Data 2022*)

Information :

K: Abundance

KR : Relative Abundance

The abundance value of bivalves at station one is 960 ind/m2 which is the second highest value after station three. The relative abundance with the highest percentage at station one was found in the type of Mactra chinensis with a percentage of 38.89%. This means that Mactra chinensis is a type of bivalves that contributes the greatest abundance value at station one. According to Kurniawan (2007) abundant species are species that can adapt to life more than other species. Mactra chinensis has a thick shell, so it can protect itself from predators or environmental conditions.

The lowest relative abundance was found in Isognomon ephippium with a percentage of 6.25%. Ephippium isognomon can attach to rocks and other hard substrates in sea and brackish water and attach to mangrove roots in estuaries and mangrove areas (Carpenter & Niem, 1998).

Based on the habitat characteristics of Isognomon ephippium, it is suspected that this species is not abundant because the mangrove area of station one has a substrate, namely muddy sand.

Mangrove density at this station is in the medium category but has the second highest abundance of bivalves among other stations. This is influenced by the habitat characteristics of the bivalves.

At station two the abundance and relative abundance of bivalves showed the lowest results from the other stations. Can be seen in Table 3.3.

Table 3.3. Data from Analysis of Abundance of Bivalves Station 2

Station		Species	(ind)	K (ind/m^2)	KR (ind)
2	1	*Mactra violacea*	1	20	8,33
		Mactra chinensis	5	100	41,67
		Geloina expansa	1	20	8,33
		Polymesoda bengalensis	5	100	41,67
	2	*Mactra violacea*	4	80	30,76
		Mactra chinensis	2	40	15,38
		Geloina expansa	2	40	15,38
		Polymesoda bengalensis	5	100	38,46
	3	*Mactra violacea*	2	100	22,22
		Geloina expansa	2	40	22,22
		Polymesoda bengalensis	5	40	55,55
		Total	34	680	300

(*Source: Primary Data 2022*)

The abundance value at station two is 680 ind/m2. The relative abundance with the highest percentage was found in Polymesoda bengalensis at 55.55%. This means that Polymesoda bengalensis is a type of bivalves that contributes the greatest abundance value at station two. Where environmental factors and differences in the characteristics of each station can affect the abundance of Polymesoda bengalensis species. This species is known to live on relatively smooth substrates (Dwiono, 2003). While the relatively low abundance was found in two species, namely Mactra violacea and Geloina expansa.

Station two has a dense mangrove density. Mangrove density affects the litter produced. Litter that comes from mangroves is (leaves, stems, fruit, twigs and so on). The litter is broken down by organisms so that it provides an energy source for bivalves (Akhrianti et al, 2014). In addition to requiring an energy source, bivalves also need a suitable habitat to live in an area, but station two is an area between the confluence of seawater and fresh water, this area has extreme and changing environmental conditions that require bivalves to adapt more extra.

At station three the abundance and relative abundance of bivalves showed the highest results from other stations. Can be seen in Table 3.4.

Table 3.4. Data Result of Analysis of Abundance of Bivalves Station 3

Station		**Species**	**(ind)**	**K (ind/m^2)**	**KR (ind)**
3	**1**	***Mactra violacea***	**8**	**160**	**20,51**
		Polymesoda erosa	**10**	**200**	**25,64**
		Geloina expansa	**9**	**180**	**23,07**
		Anadara antiquata	**6**	**120**	**15,38**
		Polymesoda bengalensis	**6**	**120**	**15,38**
	2	***Mactra violacea***	**7**	**140**	**18,42**
		Polymesoda erosa	**10**	**160**	**26,31**
		Geloina expansa	**8**	**200**	**21,05**
		Anadara antiquata	**7**	**140**	**18,42**
		Polymesoda bengalensis	**6**	**120**	**15,78**
	3	***Mactra violacea***	**8**	**160**	**19,51**
		Polymesoda erosa	**9**	**180**	**21,95**
		Geloina expansa	**10**	**200**	**24,39**
		Anadara antiquata	**7**	**140**	**17,07**
		Polymesoda bengalensis	**7**	**140**	**17,07**
		Total	**118**	**2.360**	**300**

(*Source: Primary Data 2022*)

Can be seen in Table 3.3. abundance value at station three is 2.360 ind/m2. The relative abundance with the highest percentage was found in the Polymesoda erosa species at

26.31%. This means that the Bivalves Polymesoda erosa contributed the greatest abundance value at station three. Polymesoda erosa is a type of bivalves that mostly live in mangrove forest areas by immersing themselves in muddy substrates (Peter and Sivatoshi, 2001). Where the muddy substrate with the highest percentage yield was found at station three of 75.2% mud with the dominant mangrove species being Avicennia alba. Avicennia alba mangroves have roots that grow scattered (Arief, 2003). Avicennia alba root can help bind sediment. According to Buckman and Brady (1982) sediment or bottom substrate contains organic matter. Organic matter is a source of nutrients that are needed by marine organisms (Riniasih, 2015).

The density at station three is included in the very dense category, where the high density of mangroves will produce high litter and organic matter needed by bivalves as a food source. This is one factor in the high abundance of bivalves at this station.

Mangrove Type and Density

Based on the results of observations of mangroves at the research site, three types of mangrove trees were found, namely: Avicennia alba, Rhizophora mucronata and Sonneratia alba.

Table 3.6. Mangrove Tree Density

Station	Mangrove Type	K (ind/ha)	KR (%)
1	*Avicennia alba*	667	51
	Rhizophora mucronata	400	31
	Sonneratia alba	233	18
	Total	1300	100
2	*Avicennia alba*	800	44
	Rhizophora mucronata	1000	56
	Total	1800	100
3	*Avicennia alba*	1467	67
	Rhizophora mucronata	733	33
	Total	2200	100

(*Source: Primary Data 2022*)

The highest mangrove density is found at station three with a density of 2200 ind/ha and includes the criteria for very dense or very dense. At station two, the mangrove density is 1800 ind/ha and includes very dense criteria. Station one, the density of mangrove trees is 1300 ind/ha and includes the medium criteria based on KEPMEN LH No. 51 of 2003.

The value of mangrove density describes the condition of the mangrove ecosystem, the condition of the mangrove ecosystem is influenced by several factors, namely community activities, land function shifts and environmental conditions in the surrounding area (Lestarina, et al 2020). Mangroves are a source of food, which comes from mangrove litter (leaves, twigs, fruit, stems, etc.) which are decomposed by bacteria and some of the litter becomes detritus particles that are used as food for bivalves (Bengen, 2001).

Relationship between Mangrove Density and Bivalves Abundance

The results of the regression analysis showed that the R2 value indicated that the effect of mangrove density on the abundance of bivalves was in the low category, namely 0.2771. This is reinforced by the statement (Akhrianti et al., 2014) that the abundance of bivalves is influenced by biotic and abiotic factors, namely environmental conditions with the carrying capacity of physical parameters (temperature, brightness, current velocity and substrate), chemical parameters (salinity pH and DO), and environmental parameters. biology (plankton). In addition to water quality, the presence, abundance and diversity of bivalves is influenced by predator predation, competition and the composition of food availability. Differences in the number of species and structure of bivalves can also be caused by pressure and natural changes. The results of simple linear regression can be seen in Figure 2.

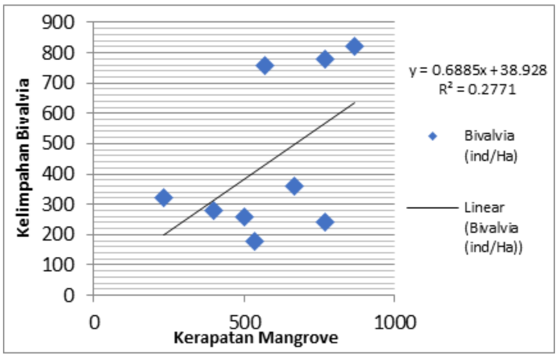

Fig.3.1 Simple Linear Regression

IV. CONCLUSION

The bivalve community in the mangrove area consists of eight species of bivalves, namely, Mactra violacea, Perna viridis, Mactra chinensis, Polymesoda erosa, Geloina expansa, Isognomon ephippium, Anadara antiquata and Polymesoda bengalensis.

The relationship between mangrove density and bivalves abundance shows that the two relationships have a relationship in the low category, but still have an influence in the low category and the rest is influenced by other environmental factors such as temperature, salinity, pH, DO and substrate.

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An Assessment of Waste Management at a Major European Based Air Cargo Terminal Operator: A Case Study of Frankfurt Cargo Services

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Abstract—*Air cargo terminal operators play a vital role in the global air cargo supply chain by acting as the key interface point between the air and surface transport modes. However, air cargo terminals produce both hazardous and non-hazardous wastes as a by-product from their operations. Using an in-depth qualitative longitudinal research design, this study has examined waste management at Frankfurt Cargo Services (FCS), one of the major European based air cargo terminal operators. The study period was from 2008 to 2019. The qualitative data was examined by document analysis. The case study found that Frankfurt Cargo Services (FCS) total annual non-hazardous wastes increased from 770 tonnes in 2009 to 1,525 tonnes in 2019. The company's hazardous wastes fluctuated over the study period from a low of 5 kilograms in 2009 to a high of 2.52 tonnes in 2010. The case study revealed that there were no reported hazardous wastes from 2014 to 2019. Frankfurt Cargo Services primary waste management method is the recovery of wastes. The annual recovered wastes increased from 770 tonnes in 2008 to 1,530 tonnes in 2019. The company's waste recovery rate increased from 95.3% in 2008 to 100% in 2019. Frankfurt Cargo Services (FCS) disposed wastes increased from 36.37 tonnes in 2008 to a high of 58 tonnes in 2017 and 58 tonnes again in 2018, respectively. There were no reported disposed wastes in 2019. During the study period, there were no reported wastes that were disposed to landfill.*

Keywords— ***Air cargo, cargo terminal operator, case study, Frankfurt Cargo Services, hazardous wastes, non-hazardous wastes.***

I. INTRODUCTION

The world air cargo industry has grown rapidly in recent decades and is now an integral part of the global economy, carrying goods valued at around $USD 6.8 trillion on an annual basis. This represents around 35 per cent of world export trade (by value) (International Air Transport Association, 2022). The air cargo supply chain is responsible for articulating the flows, both physical and documentary, of air cargo consignments from their origin to their destination (Larrodé et al., 2018). One of the very important actors in the air cargo supply chain is the air cargo terminal operator (Caves, 2015; Chen et al., 2008; Chen & Chou, 2006; Rong & Grunow, 2009). For the global movement of air cargo from an airport to an airport, the air cargo terminal is a key success factor for the terminal operator's client airlines, and hence, for the quality of air cargo transportation provided (Rodbundith et al., 2019). Air cargo terminals are facilities in which individual air cargo consignments are processed into cargo loads ready for loading onto an airline's aircraft and, following transport to their destination, are broken down again into individual shipments for delivery to the ultimate customer (Chinn & Vickers, 1998).

Waste management and the disposal of wastes are now regarded as being amongst the most important issues in the environmental management of the global airline industry

(Baxter, 2020; Li et al., 2003). Like passenger and air cargo carrying airlines, air cargo terminals also generate both hazardous and non-hazardous wastes. According to El-Din M. Saleh (2016, p. 4), "hazardous wastes are classified as hazardous if they exhibit one or more of ignitability, corrosivity, reactivity, or toxicity".

The aim of this study is to examine how a major European-based air cargo terminal operator manages its non-hazardous and hazardous wastes. A further aim of the study is to examine the annual volumes of hazardous and non-hazardous waste and the annual recovery rates of a major European-based air cargo terminal.

One such major air cargo terminal operator that has sustainably managed their air cargo handling operations is Frankfurt Cargo Services GmbH, Frankfurt Airport's largest neutral air cargo handling agent (Frankfurt Cargo Services, 2021). Since 1999, Fraport AG, a major shareholder in Frankfurt Air Cargo Services GmbH, has been regularly audited and validated by government accredited and inspected environmental auditors. Frankfurt Air Cargo Services GmbH has also been included in these environmental audits and accreditation. As such, Frankfurt Cargo Services GmbH was selected as case company for the study. A further factor in selecting Frankfurt Cargo Services GmbH as the case firm was the readily available case documentation which allowed for the in-depth analysis of the company's waste management. The study period is from 2008 to 2019.

The remainder of the paper is organized as follows: the literature review is presented in Section 2, and this sets the context for the in-depth case study. Section 3 describes the study's research methodology. Section 4 presents the case study based on Frankfurt Air Cargo Services GmbH waste management. Section 5 presents the study's conclusions.

II. BACKGROUND

2.1 The Role and Functions of an Air Cargo Terminal

Air cargo terminals serve as a temporary storage facility before the next operation can be performed, that is, loading the consignment onto its assigned flight (van Oudheusden & Boey, 1994). The cargo terminal operator (CTO) provides the handling facilities necessary to accept air cargo consignments from international air freight forwarders and shippers; check shipment weights; and prepare aircraft load plans. They also store consignments until they are cleared for export by the customs authority. The cargo terminal operator (CTO) then arranges for the air cargo consignment to be loaded onto the designated aircraft (Damsgaard, 1999). At the destination airport, an air cargo terminal operator accepts cargo from incoming flights and stores the cargo consignments until they have been cleared and released by the receiving country's customs authority. A freight forwarder then typically thereafter collects the consignment and arranges delivery to the end-customer (Martin Jones, 2013). Domestic cargo requires no customs clearance and proceeds directly from the check-in area to a pre-delivery holding area within the terminal, where it is stored pending arrangement of delivery to the final customer (Ashford et al., 2011).

The air cargo terminal provides for three principal cargo handling activities: (1) the import activities (for example, arrival of the cargo load from the aircraft, breakdown, storage of cargo pending delivery, cargo retrieval and cargo delivery), (2) the export activities (for example, unloading the cargo from customer's trucks, export cargo acceptance, export cargo handling, build-up of cargo, flight processing, retrieval of loaded aircraft unit load devices (ULDs) and cargo assembly), and (3) the transfer activity (arrival of cargo, transfer cargo handling, build-up of transfer cargo, retrieval of ULDs and cargo assembly (Ashford et al., 2011; Chen, 2004). Aircraft unit load devices, or ULDs, are pallets and containers which are used to carry air cargo, mail and passenger baggage on wide-body passenger and freighter aircraft (Baxter & Kourousis, 2015; Lu & Chen, 2011). Prior to air cargo being moved to the aircraft for departure, it is delivered to the airport to an air cargo terminal by trucks where it is then unloaded for inspection, information verification, sorting, and packing (Rodbundith & Sopadang, 2021). At the airport, the airline's cargo terminal provider (in-house or outsourced) receives the goods and documentation from the shipper, air freight forwarder, or logistics services provider. Following inspection, the freight and verifying that it is ready for air carriage, the handling company (air cargo terminal operator) loads the aircraft containers (ULDs) and builds pallets (that is, consolidates items onto aircraft pallets), and once these ULDs and pallets have been loaded they are delivered to the aircraft, where they are loaded onto the aircraft for uplift to their destination (Popescu et al., 2010).

Another major function of an air cargo terminal is information processing (Hu & Huang, 2011). Air Cargo terminal operator's computer systems are typically interface with the National Customs Administration (thus allowing the electronic clearance of air cargo consignments). Air freight forwarders and the client airlines systems are also often linked the cargo terminal operator's computer system. Prior to being exported, international air cargo consignments must be cleared for export by the relevant Customs Authority. On arrival at its destination, the air cargo consignment will undergo the relevant customs clearance formalities (Martin Jones, 2013).

An air cargo terminal can be divided into two main systems: the landside and the airside. Landside operations deal with the interchange of air cargo between air freight forwarders and logistics operators and the airport's ground handlers (GH), which receive cargo from the landside, sort the cargo and then deliver it to the corresponding aircraft (the airside) for uplift to its destination (Romero-Silva & Mota, 2018). Landside operations in air cargo terminals are comprised of many freight forwarders delivering and collecting cargo at the air cargo terminal's loading docks (Romero-Silva & Mota, 2022). Air cargo terminals handling international air cargo consignments are divided into an import area and an export area (Laniel et al., 2011: Senguttuvan. 2006). These air cargo terminals may also include an area dedicated for transfer or transshipment air cargoes (Han & Chang, 2015). An important source of air cargo for airlines is transshipment cargo, which is air cargo that is uplifted from its point of origin to its final destination via an intermediate hub airport (Merkert & Alexander, 2018).

2.2 Services Provided by Air Cargo Terminal Operators

Specialized air cargo handling firms offer a range of services from cargo warehousing through to trucking (Morrell & Klein, 2018).

The services offered by cargo handling firms include:

1. Warehousing

   Freight acceptance, build-up, and storage

   ULD build-up and breakdown

   Shipment inventory control

   Truck loading and unloading

   Express handling services

   Security services
2. Documentation
3. Handling of dangerous goods, live animals, perishables, and other special cargoes
4. Transport to and from the aircraft
5. Trucking (road feeder services) (Morrell & Klein, 2018, p. 168).

The International Air Transport Association (IATA) Standard Ground Handling Agreement (SGHA) defines the menu of services that will be offered to clients including general cargo and postal mail handling, document handling, customs control, and the handling of cargo irregularities and ramp services (Morrell & Klein, 2018, p. 168).

2.3 Types of Air Cargo Terminal Operators

An air cargo terminal operator (CTO) may be both a facility in which air cargo consignments are accepted, stored, and loaded or built up ready for air transportation, as well as the company that provides these air cargo handling services. In some instances, the air cargo handling services may be provided another firm operating within the cargo terminal. In such cases, they may be referred to as a Ground Service Provider (GSP). Air cargo terminal operators vary in size from the large multi-national firms, for example, Swissport and Worldwide Flight Services to a smaller operator that may only have operations at a single airport. The size and degree of sophistication may differ based on location as well as the annual air cargo tonnage (Donnison, 2018).

The providers of cargo handling services can be airlines (self-handling), one airline for another, airport authorities, or independent specialist ground handling firms that obtain a license to operate on the airport's facility. Airlines often service their own cargo (self-handling) and may also provide this service to other airlines (third party handling) (Morrell & Klein, 2018).

2.4 Waste Management Hierarchy

According to the Organisation for Economic Development (2003), "waste refers to materials that are not prime products (that is, products produced for the market) for which the generator has no further use in terms of his/her own purposes of production, transformation, or consumption, and of which he/she wants to dispose". The waste management hierarchy ranks the various types of wastes disposal methods from the most to the least desirable (Davies, 2016; Pitt & Smith, 2003). The waste management hierarchy is as follows: reduce, re-use, recycle, recovery, and disposal (Figure 1) (Davies, 2016; Okan et al., 2019). For firms using the hierarchy, reducing waste should be their primary concern (Baxter et al, 2018). In an ideal situation, waste should be avoided wherever possible. This means that in the waste management hierarchy, reducing or preventing waste should be the primary objective of the firm (Baxter & Srisaeng, 2021).

The waste management hierarchy seeks to minimize the generation of wastes in the first instance. The aim of the hierarchy is for the firm to optimize the opportunities for reuse and recycling of materials, and to minimize the quantities of wastes that need to be disposed to landfill (Thomas & Hooper, 2013). According to the waste management hierarchy, re-use and recycling are the best methods of dealing with unavoidable waste (Pitt & Smith, 2003). Re-using waste, wherever possible, is regarded as more favorable than recycling because the waste items does not require further processing prior to being used again (Güren, 2015). Reuse of wastes occurs when something that has already achieved its original function is once again used for another purpose. The recycling of wastes involves the reprocessing of used materials that would otherwise be considered as waste (Zhu et al., 2008). Recycling of wastes involves the collection, sorting, processing, and their conversion into raw materials that can be used in the production of new products (Park & Allaby 2013). Recovery relates to the recovery of energy that can be recovered from waste (Zhu et al., 2008). Wastes that are regarded as unsuitable for reuse or recycling can be incinerated to generate heat or electricity (Makarichi et al., 2018; Waters 2020; Zhu et al., 2008). Finally, disposal in landfill sites is regarded as the least desirable option (Manahan, 2011; Okan et al., 2019; Williams, 2013). Waste that is disposed to landfill and open dumping, is environmentally unsafe due to the emission of greenhouse gases (GHGs) that are produced from the disposed wastes (Ahmed et al., 2020; Trabold & Nair, 2019).

Fig.1: The Waste Management Hierarchy

2.5 Production of Waste in Air Cargo Terminals

In providing air cargo handling services, cargo terminal operators generate various types of waste which includes tyres, fluids from equipment, universal wastes (light bulbs, electronics, and batteries), wood and wooden pallets as well as plastic packing material (Federal Aviation administration, 2013). Table 1 shows the distinct types of waste that are typically generated at an air cargo terminal.

Table 1 – Types of wastes generated in the air cargo industry

Stakeholder	Types of waste generated
Airlines	Paper Toner cartridges Light bulbs Batteries Plastic bottles and cans Food and general rubbish
Cargo Terminal Operator	Paper Toner cartridges Light bulbs Batteries Plastic bottles and cans Food and general rubbish Green waste from landscaping activities Plastic Tyres Wood/wooden pallets
Clients	Paper Toner cartridges Light bulbs Batteries Plastic bottles and cans Food and general rubbish
Government Agencies	Paper Toner cartridges Light bulbs Batteries Plastic bottles and cans Food and general rubbish

Source: adapted from Federal Aviation Administration (2013).

In addition to the general and food waste generated from offices, other significant sources of waste are plastic packing material and wood and wooden pallets (Federal Aviation Administration, 2013). To protect air cargo consignments from the elements, plastic is used to line the base of aircraft pallets and to cover the loaded cargo on the pallet. In addition, the base of structural air cargo unit load devices (ULDs) is also often lined with plastic to protect the contents of the container. Cargo terminal operators also often shrink wrap consignments on an industrial pallet to help prevent them from moving during the transportation process. At the destination, during the unloading and handling process, the cargo terminal operator (CTO) removes the plastic shrink wrapping so they can acquit the cargo consignments and make them ready for delivery to the consignee or their appointed freight forwarder or customs agency.

For aircraft safety purposes, it can be necessary for wood to be used to spread the weight of a heavy piece cargo on an aircraft pallet to ensure the assigned aircraft's maximum floor bearing weight is not exceeded (heavy cargo shoring). Also, in the air cargo industry, wooden pallets are placed on the base of aircraft pallets, when it is necessary to raise the height of cargo that exceeds the width of the base of the aircraft fuselage (over-hang) so that the consignment can fit within the curvature of the aircraft hold. When shipping goods by air, it is necessary for the consignment to be suitably packaged. The packaging needs to be able to withstand various storage, transit, and handling conditions throughout the transportation cycle, whilst also protecting the cargo. In addition, the packaging must comply with the shipping regulations of the countries of origin and destination. The packaging used also needs to satisfy airline packaging requirements. The use of wooden packaging is very common in the air cargo industry, especially for machinery. Once the customs clearance formalities are completed at the destination airport, the consignee (or their appointed freight forwarder or customs agency), may request the CTO to remove the outer wooden packaging so that they can take delivery of the machinery or the product being shipped – this, off course, generates wooden waste. Other types of wood waste come from the use of wooden pallets used by shippers or the origin CTO for loading air cargo consignments.

III. RESEARCH METHODOLOGY

3.1 Research Approach

The study's qualitative analysis was based on a longitudinal case study design (Derrington, 2019; Hassett & Paavilainen-Mäntymäki, 2013; Neale, 2019). The key advantage of a qualitative longitudinal research design is that it reveals change and growth in an outcome or phenomena over time (Kalaian & Kasim, 2008). A case study also allows for the exploration of complex phenomena (Remenyi et al., 2010; Yin, 2018). A case study also enables the researcher(s) to collect rich, explanatory information (Ang, 2014; Mentzer & Flint, 1997).

3.2 Data Collection

The qualitative data gathered for this study was obtained from Fraport AG's annual environmental and the annual abridged environmental statements. Hence, in this study, secondary data was used in the case study analysis. The study followed the recommendations of Yin (2018) in the data collection phase, that is, the study used multiple sources of case evidence, the data was stored and analyzed in a case study database, and there was a chain of case study evidence.

3.3 Data Analysis

The qualitative data collected was examined using document analysis. Document analysis is frequently used in case studies and focuses on the information and data from formal documents and company records (Grant, 2019; Oates, 2006; Ramon Gil-Garcia, 2012). In a case study existing documents are a critical source of qualitative data, and these documents may be publicly available or private in nature (Woods & Graber, 2017). The documents collected for the present study were examined according to four criteria: authenticity, credibility, representativeness, and meaning (Fitzgerald, 2012; Fulcher & Scott, 2011; Scott, 2014).

The key words used in the database searches included "Fraport AG environmental management framework", "Frankfurt Cargo Services wastes regulatory framework", "Frankfurt Cargo Services annual hazardous wastes", "Frankfurt Cargo Services annual non-hazardous wastes", "Frankfurt Cargo Services annual disposed wastes", "Frankfurt Cargo Services annual recovered wastes", "Frankfurt Cargo Services annual wastes recoverability ratio", and "Frankfurt Cargo Services wastes handling methods".

The study's document analysis was conducted in six distinct phases. The first phase involved planning the types and required documentation and ascertaining their availability for the study. In the second phase, the data collection involved sourcing the documents from Fraport AG and developing and implementing a scheme for managing the gathered documents. In the third phase, the documents were examined to assess their authenticity, credibility and to identify any potential bias in them. In the fourth phase, the content of the collected documents was carefully examined, and the key themes and issues were identified and recorded. The fifth phase involved the deliberation and refinement to identify any difficulties associated with the documents, reviewing sources, as well as exploring the documents content. In the sixth and final phase, the analysis of the data was completed (O'Leary, 2004). The documents were all in English. Each document was carefully read, and key themes were coded and recorded in the case study (Baxter, 2021; Baxter & Srisaeng, 2021).

IV. RESULTS

4.1 Frankfurt Cargo Services GmbH: A Brief Overview

Frankfurt Air Cargo Services have been providing air cargo handling services for more than 50 years. The company has a modern air cargo terminal at Frankfurt Airport. The company has two warehouses occupying an area of 52,000 m² (Fraport AG, 2022a). The cargo terminal is connected to the apron area and there is a 100-metre distance between the freighter aircraft parking positions and the air cargo terminal (Fraport AG, 2022b). An airport's apron area is the location where aircraft stands interface with airport terminal buildings, and they are the location where aircraft are handled whilst on the ground in between flights (Budd & Ison, 2017). The company handles around 40 airlines, which includes airlines operating dedicated freighter to Frankfurt Airport. In addition, Frankfurt Cargo Services (FCS) provides the air cargo handling services for man fifty international airlines (Frankfurt Cargo Services, 2022).

On November 2, 2015, Fraport AG and Worldwide Flight Services (WFS) formed a strategic air cargo handling partnership agreement at Frankfurt Airport. Under the terms of the agreement, which was signed in July 2015, Fraport AG sold a 51 percent share in Fraport Cargo Services GmbH (FCS) to WFS (Fraport AG, 2015; Worldwide Flight Services, 2015).

Figure 2 presents the total annual air cargo tonnages handled by Frankfurt Air Cargo Services (FCS) from 2008 to 2019 together with the year-on-year change (%). The air cargo industry is extremely cyclical in nature (Oedekoven, 2010; Reynolds-Feighan, 2017; Wittmer & Bieger, 2011). This cyclicality is demonstrated in the annual tonnages of air cargo handled by Frankfurt Cargo Services. As can be observed in Figure 2, there was a pronounced spike in handled air cargo tonnages in 2010 (+35.31%). World air cargo traffic grew in 2010 (Abeyratne, 2018). This growth reflected the recovery from the 2008 and 2009 global financial crisis, which resulted in a downturn in world air cargo demand. Figure 2 shows that the annual air cargo tonnages declined on a year-on-year basis in 2011, 2012, and 2013 before returning to positive growth from 2015 to 2017. The annual air cargo tonnages decreased on a year-on-year basis in 2018 (-7.80%) and 2019 (-6.56%), respectively (Figure 2). World air cargo traffic fell quite significantly in 2019. In 2019, the air cargo industry recorded its weakest air cargo traffic performance since the global financial crisis in 2009 (International Air Transport Association, 2020).

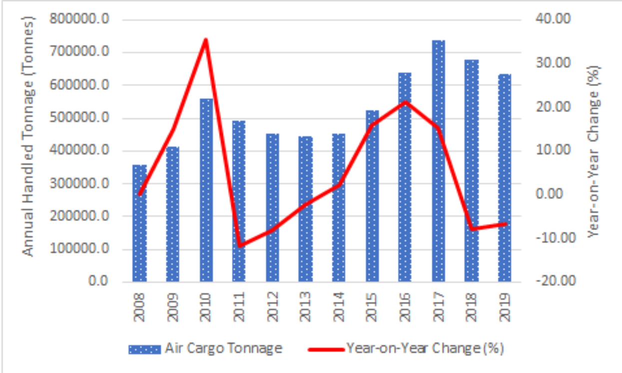

Fig.2: Frankfurt Cargo Services Annual Handled Air Cargo Tonnages and Year-on-Year Change (%): 2008-2019

Source: Data derived from Fraport AG (2012, 2014, 2018, 2019, 2020)

4.2 Fraport AG Environmental Management Framework

As previously noted, Fraport AG has held a shareholding in Frankfurt Cargo Services and, as such, Frankfurt Cargo Services has been included in Fraport AG's environmental management system (EMS).

From 1999 onwards, Fraport AG, as the manager and operator of Frankfurt Airport, has been regularly validated by government accredited and inspected environmental management auditors. The basis for such audits is the European regulation "Eco-Management and Audit Scheme" (EMAS) (Fraport AG, 2019). EMAS is a voluntary instrument of the European Union, which enables firms of any size and industry to examine and continuously enhance their environmental performance (International Airport Review, 2014). Since 2002, Frankfurt Airport's environmental audits have been carried out in compliance with the international standard ISO 14001 (Fraport AG, 2019). ISO 14001 is a global meta-standard for implementing Environmental Management Systems (EMS) (Heras-Saizarbitoria et al., 2011; Laskurain et al., 2017; Liu et al., 2020). The ISO 14001 Environmental Management System (EMS) has developed over time into one of the most widely used systems for managing corporate environmental aspects (Oliveira et al., 2011). Fraport AG's environmental audits, which comply with EMAS and ISO 14001 standards, also include the following Fraport AG subsidiaries: Fraport Cargo Services GmbH (FCS) since 2008, N*ICE Aircraft Services & Support GmbH (N*ICE) since 2009, and Energy Air GmbH since 2014 (Fraport AG, 2019).

4.2 Frankfurt Cargo Services GmbH Waste Related Regulatory Framework

Within Europe, the disposal of wastes is governed by various European regulations and directives. The European regulations automatically apply to each of the member states, whilst the directives must be separately transposed into national law by each member state. The basis of this legal framework is the Waste Framework Directive (2008/98/EC). This framework defines the key waste-related terms, lays down a five-step waste hierarchy, and contains key provisions for German waste disposal law (Umwelt Bundesamt, 2021).

Germany's first uniform national waste disposal act, the Abfallbeseitigungsgesetz (AbfG), was adopted in 1972. The Waste Management Act (KrWG), which is the current German main waste disposal statute (and the successor to the KrW-/AbfG act), incorporates the main structural elements of the Kreislaufwirtschafts- und Abfallgesetz (KrW-/AbfG). The disposal of specific types of product waste (respectively end-of-life vehicles, used batteries and end-of-life electronic and electrical devices) is governed by the ELV regulation (AltfahrzeugV), Batteriegesetz (BatterieG) and Elektro- und Elektronikgerätegesetz (ElektroG) (Umwelt Bundesamt, 2021).

Germany's Waste Management Act (KrWG) came into effect on 1 June 2012. The KrWG, which was enacted as Article 1 of the law titled "Gesetz zur Neuordnung des Kreislaufwirtschafts- und Abfallrechts", superseded the law titled Kreislaufwirtschafts- und Abfallgesetz (KrW-/AbfG) and transposes Directive 2008/98/EC into German law. The Waste Management Act (KrWG) is intended to tighten resource, climate, and environmental protection regulations (Umwelt Bundesamt, 2021).

One of the core provisions of Germany's Waste Management Act (KrWG) is the five-step (previously three step) hierarchy pursuant to Article 6, according to which the following ranking of waste management measures applies:

- Prevention
- Preparation for recycling
- Recycling
- Other types of recovery, particularly use for energy recovery
- Disposal (Umwelt Bundesamt, 2021)

4.4 Frankfurt Cargo Services GmbH Annual Wastes

Frankfurt Air Cargo Services (FCS) total annual wastes and the year-on-year change (%) from 2008 to 2019 are presented in Figure 3. As can be observed in Figure 3, the company's total annual wastes oscillated over the study period. There was a general upward trend from 2008 to 2010, when the annual wastes increased from 770 tonnes in 2008 to 1,120 tonnes in 2010. This was followed by a general downward trend from 2011 to 2013, with total wastes decreasing from 1,.080 tonnes in 2011 to 900 tonnes in 2013. From 2014 to 2017, there was again a general upward trend in the company's annual wastes. The highest amount of annual waste was recorded in 2017 (1,668 tonnes). Figure 5 shows that there were five years in the study period where the company's annual wastes increased on a year-on-year basis. These annual increases occurred in 2009 (+10.38%), 2010 (+31.76%), 2014 (+5%), 2015 (+0.1%), 2016 (+37.73%), 2017 (+28.01%), respectively (Figure 3). In each of these years, Frankfurt Cargo Services (FCS) handled increased volumes of air cargo, and this resulted in higher amounts of waste in these respective years. In 2009, the company was able to handle a 15.08% increase in its air cargo traffic, whilst at same time only experiencing an increase of 10.38% in its annual wastes. A similar situation occurred in 2010, when the company handled an increase of 35.31% in its air cargo traffic, yet annual wastes increased at a slightly lower annual rate of 31.76%. In 2014, the company increased its annual air cargo traffic by 2.16% but its wastes increased by 5%, which indicates that the annual waste volumes grew at a slightly higher rate than that of the air cargo traffic growth rate. In 2015, the company increased its annual air cargo traffic throughput by 15.97%, whilst at the same time its annual wastes increased by just 0.1%. This was a very favorable result and showed that the company was able to handle greater air cargo volumes whilst at the same time limiting its annual wastes. There was a slightly different situation in 2017 and 2018, when the annual wastes increased at a higher rate than the annual air cargo traffic growth rate, reflecting a different waste pattern in both 2017 and 2018. As can be observed in Figure 3, there were five years in the study period where Frankfurt Cargo Services (FCS) annual wastes decreased on a year-on-year basis. These annual decreases occurred in 2011 (-3.57%), 2012 (-9.35%), 2013 (-8.06%), 2018 (-0.05%), and 2019 (-8.51%), respectively (Figure 3). In each of these years, Frankfurt Cargo Services (FCS) handled less air cargo traffic, and, as a result, the lower levels of air cargo traffic handled resulted in smaller quantities of wastes generated. Its is important to note that solid wastes can be heterogenous in nature (Abdel-Shafy & Mansour, 2018; Norbu et al., 2005; Perazzini et al., 2016), and hence, wastes can fluctuate in line with a firm's activities and processes. Furthermore, air cargo is heterogenous in nature (Balliauw et al., 2016; Kupfer et al., 2011; Otto, 2017). In addition, cargo terminals experience variations in their air cargo flows and air cargo flows may vary by airline

(Ashford et al., 2013). Air cargo comes in all shapes, sizes, weights, and packaging and this heterogeneity can influence the amount of waste that is produced in handling air cargo traffic.

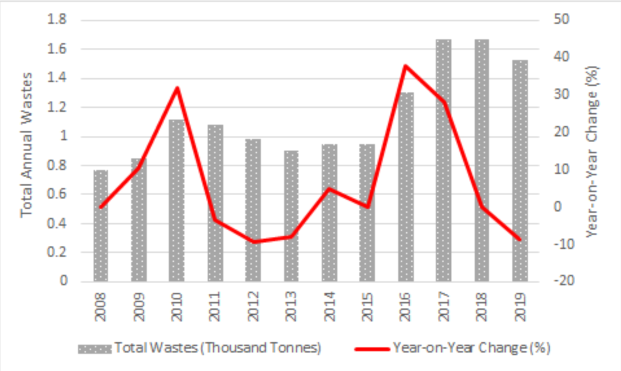

Fig.3: Frankfurt Cargo Services Total Annual Wastes and Year-on-Year Change (%): 2008-2019

Source: Data derived from Fraport AG (2012, 2014, 2018, 2019, 2020)

Figure 4 presents Frankfurt Cargo Services annual wastes per workload unit (WLU) and the year-on-year change (%) for the period 2008 to 2019. One workload (WLU) or traffic unit is equivalent to 100 kilograms of air cargo handled (Doganis, 2005; Graham, 2005; Teodorović & Janić, 2017). As can be observed in Figure 4, there are two discernible trends with this metric. From 2008 to 2015, there was a general downward trend in this metric, with the annual wastes per workload unit (WLU) decreasing from a high of 0.241 kilograms per workload unit (WLU) in 2008 to a low of 0.180 kilograms per workload unit (WLU) in 2015. From 2016 to 2019, there was an upward trend in the annual wastes per workload unit (WLU). The highest annual wastes per workload unit (WLU) was recorded in 2018 (0.246 kilograms per workload unit WLU). Figure 4 shows that the largest single annual decrease in this metric occurred in 2009, when the company's total annual wastes per workload unit (WLU) decreased by 14.52% on the 2008 levels. This may have been due to the very strong growth in handled tonnage in 2008, which was considerably higher than the annual growth in annual wastes in that year. That is, the company handled more air freight traffic, and thus, had a higher throughput to spread the generated wastes over. There was a further significant annual decrease in this metric in 2015, at which time it decreased by 13.87% on the 2014 levels (Figure 4). In 2015, Frankfurt Cargo Services (FCS) was able to handle higher volumes of air cargo, which grew at a higher rate than the associated wastes growth rate. Thus, the company was able to spread the total wastes over more workload units (WLU), thereby lowering the value per workload unit (WLU) in 2015. The significant decreases in this metric in 2009 and 2015 are a favorable result for the company as they were able to handle larger amounts of air cargo, which increased at a higher rate than the company's waste rate. Figure 4 shows that there was a pronounced spike in this metric in both 2016 (+13.33%), and 2017 (+11.27%), respectively. These two spikes in this metric could be attributed to the increase in the company's wastes being higher than the growth in its annual air cargo volumes handled. This trend led to a situation where there were fewer workload units (WLUs) to proportion the total wastes over in both 2016 and 2017, and thus, there was an increase in the annual wastes per workload (WLU) unit in both these years.

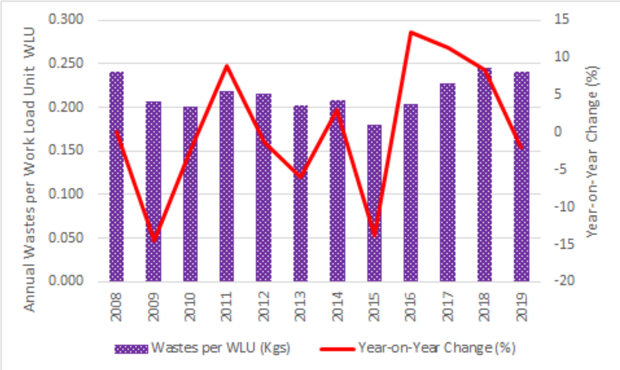

Fig.4: Frankfurt Cargo Services Total Annual Wastes Per Workload Unit (WLU) and Year-on-Year Change (%): 2008-2019

Source: Data derived from Fraport AG (2012, 2014, 2018, 2019, 2020)

Frankfurt Cargo Services (FCS) total annual hazardous wastes and the year-on-year change (%) for the period 2008-2019 are presented in Figure 5. As can be observed in Figure 5, once again there were two discernible trends with the annual amount of these wastes. Firstly, the company handled hazardous wastes in the period 2008 to 2013, whilst from 2014 to 2019 there were no reported hazardous wastes handled by the company. Figure 5 shows that there was a very significant spike in the amounts of hazardous wastes in 2010, when they increased from 5 kilograms in 2009 to 2,523 kilograms in 2010. Figure 5 also shows that there was another significant increase in these wastes in 2012, when they increased by 99.35% on the 2011 levels. There were two years in the study period where the company's hazardous wastes decreased on a year-on-year basis. These decreases were recorded in 2011 (-93.89%), and 2013 (-21.82%), respectively, and reflect the lower demand for the air transportation of hazardous cargoes in both 2011 and 2013. The air cargo sector is highly cyclical in nature (Manners-Bell, 2017), and the hazardous air cargoes handled by Frankfurt Cargo Services (FCS) have appeared to be cyclical in nature.

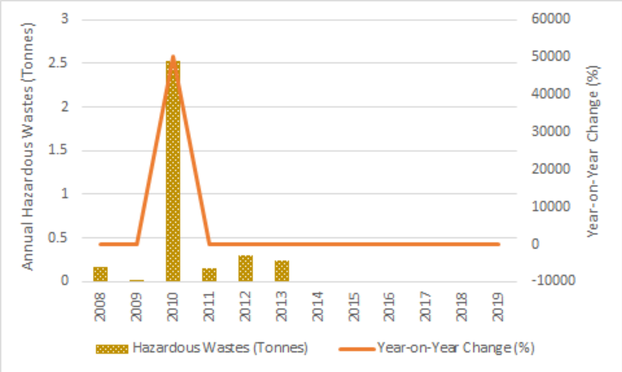

Fig.5: Frankfurt Cargo Services Annual Hazardous Wastes and Year-on-Year Change (%): 2008-2019

Source: Data derived from Fraport AG (2012, 2014, 2018, 2019, 2020)

Frankfurt Cargo Services (FCS) total annual nonhazardous wastes and the year-on-year change (%) for the period 2008-2019 are presented in Figure 6. As can be observed in Figure 6, Frankfurt Cargo Services (FCS) total annual non-hazardous wastes have largely exhibited a general upward trend throughout the study period, increasing from 770 tonnes in 2008 to 1,525 tonnes in 2019. This general upward trend is demonstrated by the year-on-year percentage change line graph, which is more positive than negative, that is, more values are above the line than below. The largest single annual increase in the company's non-hazardous wastes was recorded in 2016, when these wastes increased by 36.84% on the previous year's levels. As previously noted, Frankfurt Cargo Services (FCS) recorded an increase in its annual wastes in 2016, which were associated with higher levels of air cargo handled by the company in 2016. Figure 6 also shows that there were four years throughout the study period when the total amounts of these non-hazardous wastes decreased on a year-on-year basis. These annual decreases were recorded in 2011 (-3.57%), 2012 (-9.9%), 2013 (-7.5%), and 2019 (-8.38%), respectively. Both Frankfurt Cargo Services (FCS) annual wastes and air cargo traffic decreased on a year-on-year basis in 2011, 2012, 2013, and 2019, and this translated into the annual decreases in non-hazardous wastes in these respective years. During the study period, non-hazardous wastes accounted for the largest share of the wastes produced by Frankfurt Cargo Services.

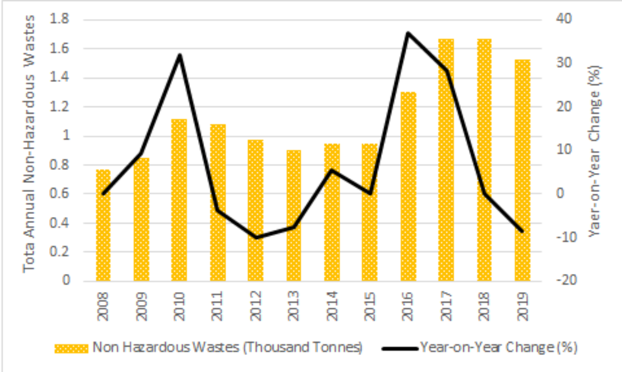

Fig.6: Frankfurt Cargo Services Annual Non-Hazardous Wastes and Year-on-Year Change (%): 2008-2019

Source: Data derived from Fraport AG (2012, 2014, 2018, 2019, 2020)

Figure 7 presents Frankfurt Cargo Services total annual disposed wastes and the year-on-year change (%) for the period 2008-2019. As can be observed in Figure 7, the company's annual disposed wastes oscillated throughout the study period. The largest single annual increase in disposed wastes was recorded in 2010, when they increased by 53.95% on the 2009 levels. In 2010, Frankfurt Cargo Services (FCS) total annual wastes and air cargo traffic both recorded strong growth on the 2009 levels, and thus, there were more wastes to be disposed of in 2010. Figure 7 shows that there were two further annual spikes in this metric during the study period. These occurred in 2016 (+19.29%) and 2017 (+21.84%), and these increases reflected the growth in the volume of air cargo handled and the associated wastes that are produced from these services. There were four years in the study when the company's disposed wastes declined on a year-on-year basis. These annual decreases occurred in 2009 (-3.68%), 2011 (-17.67%), 2012 (-16.66%), and 2014 (-1.81%), respectively. In 2009, the company produced more waste, but the types of waste that required disposal declined on a year-on-year basis, which was a favorable outcome. In 2012 and 2014, the quantity of wastes generated and the types of waste requiring disposal as the waste handling method decreased on a year-on-year basis, and once again, this was a favorable outcome for the company. In 2014, the company generated more waste, but the types of waste that required disposal decreased on a year-on-year basis Figure 7 shows that the annual disposed wastes remained constant in 2017 and 2018 at 58 tonnes. There were no disposed wastes recorded in 2019.

Since 2005, Germany has established very high criteria for the operation of landfill sites. Consequently, from 2005 onwards, Frankfurt has ceased to use landfill sites for the disposal of wastes (Frankfurt Green City, 2011). During the study period, there were no reported wastes that were disposed to landfill.

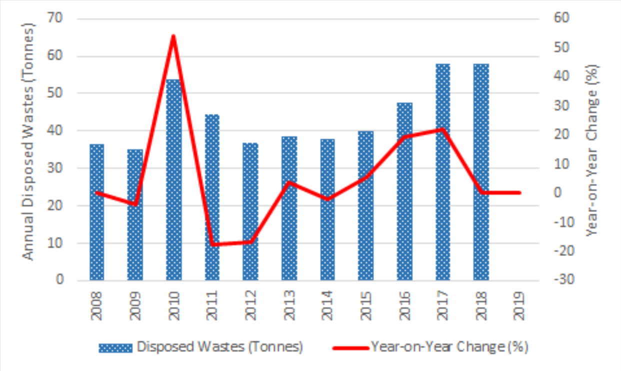

Fig.7: Frankfurt Cargo Services Annual Disposed Wastes and Year-on-Year Change (%): 2008-2019

Source: Data derived from Fraport AG (2012, 2014, 2018, 2019, 2020)

Frankfurt Cargo Services (FCS) primary environmentally sustainable waste handling method is the recovery of wastes. Frankfurt Cargo Services total annual recovery wastes and the year-on-year change (%) from 2008-2019 are presented in Figure 8. As can be observed in Figure 8, Frankfurt Cargo Services annual recovered wastes has principally displayed an upward trend, increasing from 770 tonnes in 2008 to 1,530 tonnes in 2019. This general upward trend is demonstrated by the year-on-year percentage change line graph, which is more positive than negative, that is, more values are above the line than below. Figure 8 shows that there was a quite pronounced increase in the annual amounts of recovered wastes in 2010 (+31.76%), 2016 (+38.46%), and 2017 (+27.77%), respectively. Figure 8 also shows that there were three years during the study period, when the annual recovered wastes decreased on a year-on-year basis. These decreases occurred in 2011 (-8.03%), 2012 (-9.02), and in 2019 (-4.96), respectively. These annual decreases reflected the lower volumes of waste produced by the company in these respective years. Figure 8 also shows that the annual recovered wastes remained constant in 2017 and 2018 at 1,610 tonnes. Overall, the amount of recovered wastes accounts for the largest share of disposed wastes by the company, and the recovery of wastes provides significant environmental benefits. The recycling or reusing wastes results in a reduction in the quantity of wastes sent for incineration, the conservation of natural resources, energy savings, and a reduction in pollution by reducing the requirement to collect new raw materials (United States Environmental Protection Agency 2020).

Fig.8: Frankfurt Cargo Services Annual Recovered Wastes and Year-on-Year Change (%): 2008-2019

Source: Data derived from Fraport AG (2012, 2014, 2018, 2019, 2020)

Figure 9 presents Frankfurt Cargo Services annual wastes recovery rate and the year-on-year change (%) for the period 2008 to 2019. As can be observed in Figure 9, the company's annual waste recovery rate has displayed an upward trajectory, increasing from 95.3% in 2008 to 100% in 2019. This overall increase is once again demonstrated by the year-on-year percentage change line graph, which is more positive than negative, that is, more values are above the line than below. Figure 9 shows that the highest single annual increase in this metric was recorded in 2019, when the company's waste recovery ratio increased by 3.52% on the 2018 ratio. There were two years in the study period where this annual ratio declined on a year-on-year basis. These declines were recorded in 2010 (-0.41%) and 2015 (-0.2%), respectively. In both 2010 and 2015, Frankfurt Cargo Services (FCS) recorded increases in the wastes that required disposal and, as such, these wastes may not have been suitable for re-use. In 2017 and 2018, the ratio remained the same, that is, 96.5% (Figure 9). The very high recovery of wastes by Frankfurt Cargo Services (FCS) is very favorable, and this helps to mitigate its impact on the environment. Material and resource recovery from waste has significant environmental benefits. Resource recovery in the form of material, energy, or fuel from waste, not only contributes directly to fulfilling and offsetting the resource demand of society, but it also saves energy, water, and avoids harmful greenhouse gas (GHG) emissions. In addition, these resources may have economic benefits (Uz Zaman, 2016).

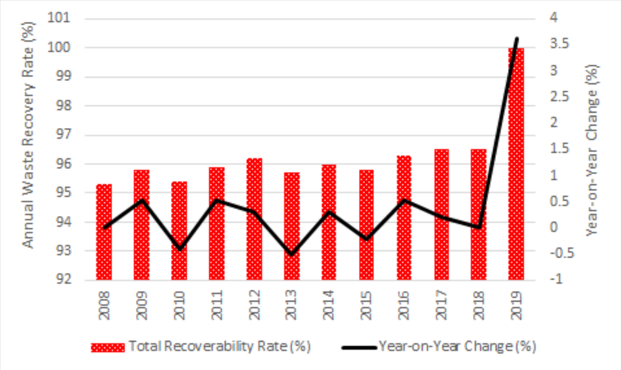

Fig.9: Frankfurt Cargo Services Annual Waste Recoverability Rate (%) and Year-on-Year Change (%): 2008-2019

Source: Data derived from Fraport AG (2012, 2014, 2018, 2019, 2020b)

V. CONCLUSION

In conclusion, this study has examined the waste management at Frankfurt Cargo Services (FCS), a major European-based air cargo terminal operator. To achieve the study's research objectives, Frankfurt Cargo Services (FCS) was selected as the case firm. The study's research was based on an in depth qualitative longitudinal research approach. The data collected for the study was analyzed by document analysis. The period of the study was from 2008 to 2019.

The case study found that Frankfurt Cargo Services (FCS) total annual non-hazardous wastes increased from 770 tonnes in 2009 to 1,525 tonnes in 2019. The company's hazardous wastes fluctuated over the study period from a low of 5 kilograms in 2009 to a high of 2.52 tonnes in 2010. The case study revealed that there were no reported hazardous wastes from 2014 to 2019.

Frankfurt Cargo Services primary waste management method is based on the recovery of wastes. The annual recovered wastes increased from 770 tonnes in 2008 to 1,530 tonnes in 2019. The company's waste recovery rate increased from 95.3% in 2008 to 100% in 2019. The recycling or reusing wastes results in a reduction in the amount of wastes that need to be disposed. Recovering wastes enable a firm to conserve natural resources, achieve energy savings, and a reduce pollution by reducing the requirement to collect new raw materials.

Frankfurt Cargo Services disposed wastes increased from 36.37 tonnes in 2008 to a high of 58 tonnes in 2017 and 58 tonnes in 2018, respectively. There were no reported disposed wastes in 2019, and, importantly, there were no reported wastes that were disposed to landfill during the study period.

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Prevalence of Cashew Powdery Mildew Disease in Western Province of Zambia

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***Abstract*— *Cashew powdery mildew disease (PMD) is the most devastating disease of cashew nuts lowering nut yields and quality in Zambia and the rest of the world. Information on prevalence patterns and timing of disease onset is vital to manage the disease in any country effectively. Cashew powdery mildew disease incidence and severity were assessed from 160 farmers randomly chosen from all agricultural camps in eight of the ten cashew-growing districts of the Western province of Zambia in April, May, June and July 2020. A two-way ANOVA was used to compare disease severity by month and district. Results showed significant differences (P < 0.001) in both PMD incidence and severity among different districts, with the highest incidence in Limulunga (55.88 %) and the least in Sikongo District (36.49 %). Disease severity was highest in Nalolo (57.49 %) and lowest in Sioma district (27.87 %). July registered the highest (61.89 %), and April had the lowest (20.08%) incidence of the disease. PMD severity was highest in July (51.69 %). The current study indicates that one of the best strategies to control PMD in this cashew-growing province of Zambia is to include control measures such as the removal of water shoots beginning February through March for all districts, chemical control beginning in April in Limulunga district and delayed to May for the rest of the districts. The study has also identified the hotspots of PMD that should be priority targets for disease management to maximize the use of limited resources.*

***Keywords*— Anacardium occidentale, powdery mildew disease, incidence, severity**

I. INTRODUCTION

Cashew (*Anacardium occidentale* Linn.) is an evergreen perennial nut crop in the flowering family *Anacardiaceae* (Majune *et al*., 2018; Ohler, 1979). It is native to northeastern Brazil but is widely grown in tropical climates, including Zambia. The plant was introduced to Africa in the 16th century (Ohler, 1979) and to Zambia in the 1940s (NA, 2016). Cashew is an important crop for nutrition and income generation worldwide (Majune *et al*., 2018; Mange *et al.,* 2014).

The current cashew yield in Zambia stands at 0. 850 MT/ha despite having a potential of 1.3 MT/ha (NA, 2016). This low yield is due to various abiotic and biotic factors that severely constrain cashew production in Zambia (ZARI, 2018). Some of the biotic humpers include pests and diseases such as *Helopeltis* and powdery mildew, respectively. Although the relative extent of damage is by far unknown, powdery mildew is probably the most devastating biotic factor in cashew production in Zambia. The disease was first detected in Zambia in 1979 (Uaciquete *et al*., 2003; Zhongrun & Masawe, 2014), and it is known to be the major constraint to cashew production worldwide (Sijaona *et al.,* 2006), associated with crop losses of 70 to 100% (Shomari & Kennedy, 1999; Sijaona & Shomari, 1987). The disease affects all young parts of the shoot, including leaves, inflorescences, apples, and nuts, reducing the visual quality and yield of cashew apples and nuts (Glawe, 2008; Sijaona & Shomari, 1987) as well as the nutritional status (Uaciquete *et al*.,2017).

Although smallholder farmers have reported the occurrence of PMD in Zambia, the extent of the damage has not been quantified (ZARI, 2018 Unpublished). In addition, incidence and severity patterns and hotspots of PMD have yet to be systematically assessed, thereby hindering the deployment of appropriate control strategies for this disease in Zambia. In addition, knowledge of the timing of disease onset in Zambia remains largely unknown, yet such information is vital in triggering the deployment of control measures. Currently, cashew farmers in Zambia use chemical control in a haphazard manner, a situation that has resulted in the excessive use of dangerous chemicals, engendering humans and the environment. Therefore, the current study sought to assess the incidence and severity of cashew powdery mildew disease to generate information for improved management of the disease in the cashew-growing region of Zambia.

II. MATERIALS AND METHODS

3.1.1. Study Locations

The survey was conducted in 8 districts of the western province, namely, Kalabo, Limulunga, Lukulu, Mongu, Nalolo, Senanga, Sikongo and Sioma (Figure 1). The districts were purposively selected because they are the cashew-growing districts of the province. Both commercial and family cashew orchards were assessed in the study.

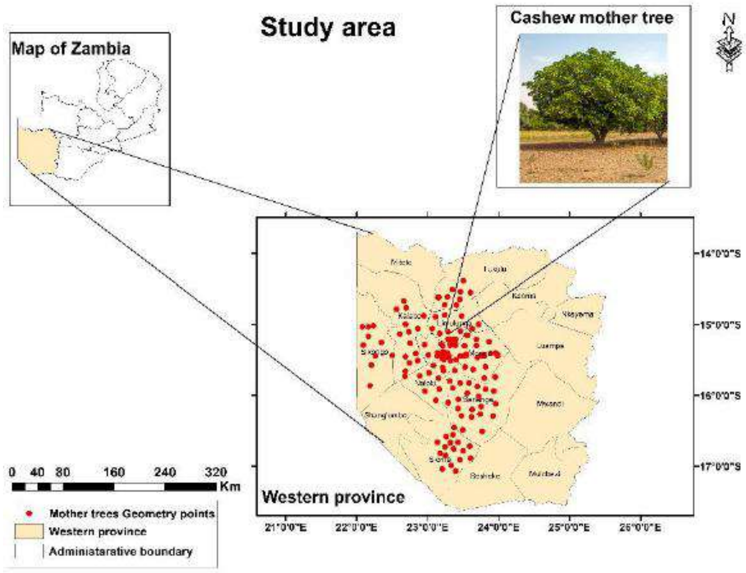


Fig.1: Sampling sites in 8 cashew-growing districts of the Western Province of Zambia.

Map developed using the software ArcGIS version 10.7.1 from the GPS coordinates recorded during the survey.

Disease incidence and severity Assessment

Disease incidence and severity were assayed on 160 trees at the first flash, flowering and panicle initiation stages of cashew from April to July 2020, as done previously (Sijaona *et al*., 2006). Depending on the size of the plantation, the zigzag or transact-walk sampling method was used to pick at least 20 cashew trees in each farmer's field (Gomez & Gomez, 1984). PMD incidence and severity were assessed per field and sampling date, based on the previously standardized scale (Nathaniels, 1990; 1996; Sijaona & Mansfield, 2001) as shown in table 1 below;

Table 1: Powdery mildew disease severity standard used in the study (modified from Nathaniels, 1990)

Infection Scale	Infection
0	No lesions
1	Powdery mildew fungus covering less than 5% of the leaf
2	Powdery mildew fungus covering 5% - 15% of the leaf
3	Powdery mildew fungus covering 15% - 25% of the leaf
4	Powdery mildew fungus covering 25% - 35% of the leaf
5	Powdery mildew fungus covering more than 35% of the leaf

The north and south sides of the tree canopy were scored based on the above standard (Table 1) using a 1 m x 1 m quadrant (Majune *et al.*, 2019). Four rounds of scoring, one month apart, corresponding with the phenological stages indicated previously (Adiga *et al*., 2019), were conducted for both PMD incidence and severity. Severity scores were converted to percentages for ease of presentation. Tree means were used to perform the statistical Analysis for PMD severity.

III. DATA ANALYSIS

Data were checked for normality, and where necessary, arcsine transformed prior to Analysis (Gomez & Gomez, 1984). However, only actual means are presented for clarity. A 4 x 8 two-way analysis of variance (ANOVA) was used to assess variability in incidence and severity among districts and months and to check for the presence of interaction between district or location and month of sampling. Means were separated using the Bonferroni test at a 5% significance level because of its advantage of allowing the comparison of several variables to avoid false data appearing statistically significant.

IV. RESULTS

Cashew Powdery mildew disease incidence across districts and months

There were significant differences ($P < 0.001$) in PMD incidence and severity among the different districts and months (table 2). The incidence and severity in districts depended on the month of assessment.

Table 2.0 Mean squares for Analysis of variance of cashew powdery mildew disease incidence and severity across districts evaluated from Western Province.

Source of variation	d.f	MS PMD Incidence	MS PMD everity
District	7	2287.5***	9081.854***
Month	3	63463.6***	41741.4***
District x Month	21	1157***	3097.08***
Error	608	113.7***	55.04***
Total	639		

*** Data significant at $P = 0.01$; d.f- degree of freedom, ms- mean square

The highest severity was recorded in Nalolo (57.48%), followed by Sikongo (50.61%), while Sioma had the lowest severity (27.84%). The district with the highest incidence was Limulunga (55.88 %, table 3), followed by Senanga (49.14%), while Sikongo had the least (36.49 %).

District	Severity (%)	Incidence (%)
Sioma	27.84[a]	47.15[b]
Limulunga	29.45[ab]	55.88[c]
Lukulu	31.53[b]	36.49[a]
Senanga	31.89[b]	49.14[b]
Mongu	37.58[c]	44.51[b]
Kalabo	38.73[c]	47.23[b]
Sikongo	50.61[d]	46.32[b]
Nalolo	57.48[e]	46.12[b]
Mean	**37.102**	**46.61**

Table 3.0: PMD severity and incidence by the district in the Western Province of Zambia in 2020

[a]Means followed by the same letter within the column are not statistically different by Fisher's protected least significant difference test performed at P = 0. 05

The highest severity was recorded in July (51.69%, table 4), followed by June (50.9%), while April had the lowest severity (18.27%). The months with the highest incidence of PMD were June (61.89%) and July (61.96%), followed by May (42.49%), while April had the least (20.08 %).

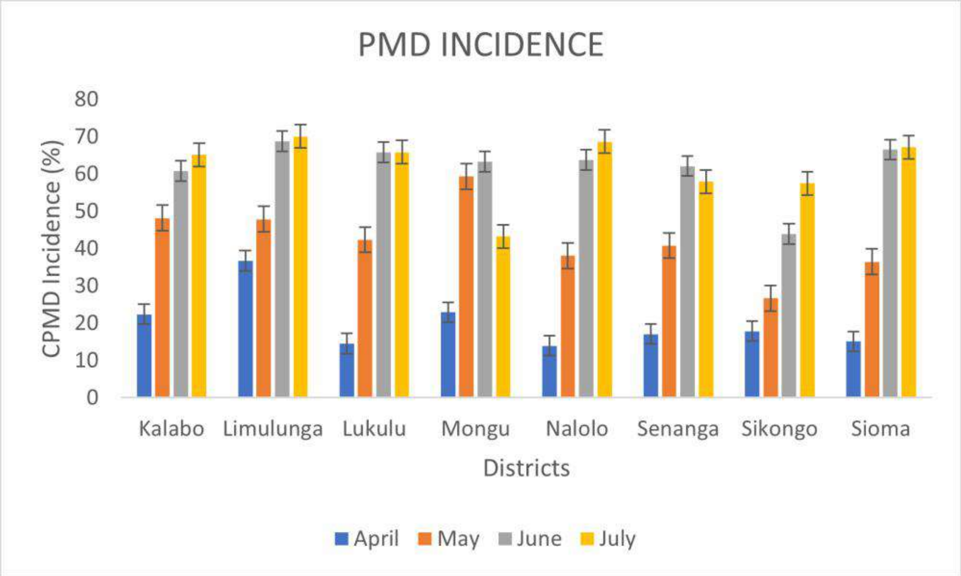
Table 4.0: PMD severity and incidence by month in the Western Province of Zambia in 2020

Months	Severity (%)	Incidence (%)
April	18.27[a]	20.08[a]
May	31.69[b]	42.49[b]
June	50.9[c]	61.89[c]
July	51.69[d]	61.96[c]
Mean	**38.14**	**46.6**

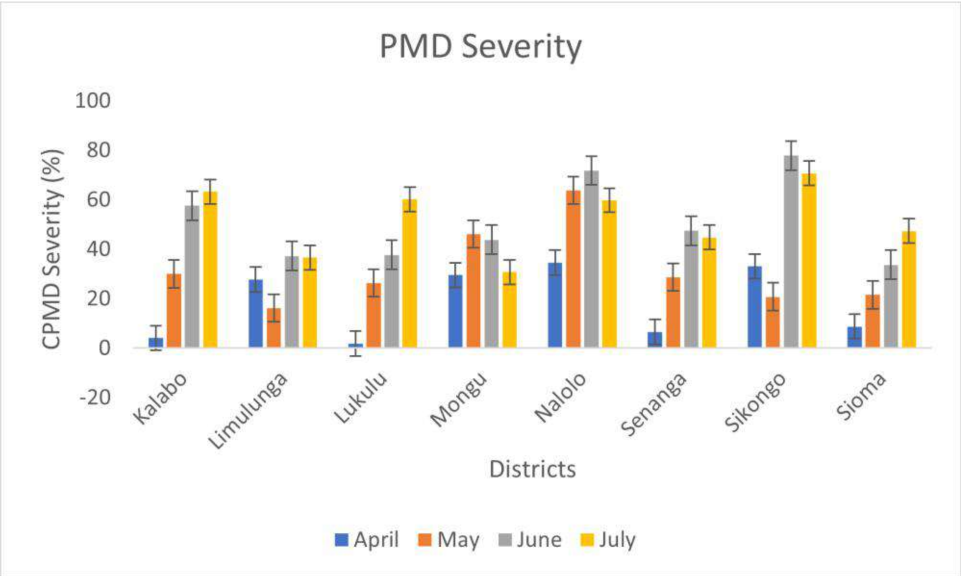
[a]Means followed by the same letter within the column are not statistically different at P=0.05

The disease incidence in the districts depended on months as there was a significant location-by-month interaction which was observed as shown in graphs 1 and 2 below;

Graph 1. PMD incidence from April to July 2020



Graph 2. PMD severity from April to July 2020



V. DISCUSSION

Prevalence of CPMD by district

This study determined the incidence and severity of cashew powdery mildew disease in the Western Province of Zambia. The results showed that Limulunga district had the highest PMD incidence (55.88%) while Sikongo district had the lowest (36.9%). These results indicate that PMD is widespread in the locations studied. These relatively high occurrences of PMD in cashew nuts could be due to the predominant use of susceptible planting materials (Wonni *et al*., 2017; Masawe, 1996) or favourable environmental conditions for developing PMD. The temperatures during study periods ranged from 15^{0}C to 28^{0}C, a range very supportive of PMD (Sijaona *et al*., 2001). In addition, inappropriate cultural practices such as lack of selective thinning, formative pruning, top-working, gap filling using grafted seedlings, clonal and polyclonal seedlings, intercropping, the possible emergence of a new and virulent pathotype, and a possible narrow genetic base of the cashew population in Zambia could be responsible for the observed high incidence and severity in the study sites (Milheiro & Evaristo, 1994; Prasad *et al*., 2000). However, studies are needed on the virulence of the causal organism in Zambia to confirm this. Although the prevalence rates were high in all districts, Limulunga has much higher rates than the rest of the province. Although the results clearly show variation in the prevalence of

PMD in the study sites and that genetic-environmental interactions may have contributed to the observed differences in powdery mildew prevalence, it is improbable that the environment had a significant impact on the incidence and severity of PMD in the study sites since the environmental conditions for the study sites do not differ significantly. The sites lie in the same agroecological zone with similar rainfall, temperature, and humidity. Therefore, such observations are more likely resulting from the effect of host susceptibility and pathogen virulence differences from one district to the next PMD (Majune *et al*., 2018; Sijaona *et al.*, 2005; Sijaona *et al.*, 2006). This study clearly shows that Limulunga should receive proportionately higher mitigation efforts compared to other district districts in order to prevent losses due to PMD in the district.

Prevalence of CPMD by month

In general, the PMD incidence and severity were very low in April when the temperatures were slightly high and increased with a reduction in temperatures from May to July, which has the coldest days or lowest temperatures in the Western Province. This could be because the epidemic's development is favoured by dry and cold conditions (Glawe, 2008; Waller *et al.*, 1992; Milheiro & Evaristo, 1994). This is supported by previous studies (Majune *et al*., 2018; Agrios, 2005; Lopez, 2008; Suffert *et al*., 2011) that showed that three factors necessary for the disease to occur included host susceptibility, pathogen virulence, and a low temperature. In addition, earlier studies (Shomari & Kennedy,1998) demonstrated that *O. anarcadii* conidial germination and cashew tissue infection by a PMD pathogen occurred over a wide range of temperatures and humidity. Therefore, environmental factors that promote the fungus's growth and viability are expected to result in higher PMD disease prevalence (Shomari & Kennedy, 1999). Furthermore, differences in weather conditions are expected over time and between various cashew production areas (Nathaniels & Kennedy, 1996). Therefore, the different district patterns of epidemic development observed in the study sites agree with the findings reported in different regions of Tanzania (Nathaniels *et al.*, 1993; Nathaniels, 1990) and Mozambique (Uaciquete *et al.*, 2003; Nathaniels, 1994; Topper *et al*., 2000).

The phenological development stages of cashew nuts that are very susceptible to PMD attack are the leaf, shoot, inflorescence, flowering, and fruit development stages (Adiga *et al.*, 2019). In Zambia, these development stages occur from April to September. From December to early April, the cashew's principal growth stage is the vegetative leaf development stage, with minimal availability of susceptible tissue (Uaciquete *et al.*, 2003; Uaciquete *et al*., 2017) and unfavourable environmental conditions (Nathaniels *et al.*, 1996), hence the lower disease prevalence observed for April in all the study sites. The higher prevalence of the disease observed in Zambia from May through to July could be attributed to a more extended period of availability of susceptible tissue, a favourable environmental condition, and a virulent pathogen (Agrios, 2005), as it coincides with the most susceptible phenological developmental stages of cashew (Adiga *et al.*, 2019). Zhai *et al.* (2021) reported that the highest combined and mixed effects of temperature variables and the duration of leaf development to the maturation of rubber PMD are strongly influenced by both extrinsic and intrinsic factors, as that found in oak trees, which are highly affected by winter temperature and phenology (Marcais *et al*., 2018). The observed PMD progress in the study areas could also be related to the inoculum density in cashew trees (Martins *et al.*, 2018). Although the present study did not investigate the effect of temperature from one month to the next and leaf development phenology on cashew PMD severity, this is very likely for cashew, as observed from previous studies (Uaciquete *et al*., 2013; Nathaniels & Kennedy, 1996; Adiga *et al.*, 2019).

Cross-cutting issues

The above observations have led to the hypothesis that chemical applications for disease mitigation require more frequency in some regions than others because of climatic differences. This suggests that disease mitigation in Sikongo, Nalolo, Mongu, and Limulunga districts should begin in mid-April and in May for the rest of the districts to manage the disease effectively. This is because the disease control severity threshold should be 20% or above before reaching the economic threshold (Sijaona *et al.*, 2001). Therefore, severe losses in both cashew nut quality and quantity would be experienced if disease control is not done by this time. Field observations showed that the dwarf cashew genotypes exhibited high levels of disease tolerance compared to the common giant genotypes in the leaf-flushing phenological stages but were both susceptible at the inflorescence and flowering stages (unpublished). This observation suggests that chemical control of the disease for the dwarf genotypes could start at the inflorescence and flowering stages for the dwarf genotypes and the leaf-flushing development stage for the common giant genotypes, as this would be more economical even for resource-limited farmers. The variation in microclimate likely plays a role in the development and the evolution of the disease in the study locations as it was associated with the types of cashew genotypes, location, and the phenology of the trees (Lopez, 2008; Suffert *et al*., 2011).

VI. CONCLUSION AND RECOMMENDATIONS

This study revealed that PMD is widespread in the area surveyed and that prevalence differed by month and district. The study also revealed that incidence and severity increased from April to July and were highest in the Limulunga district. Therefore it is recommended that control measures such as the removal of water shoots should begin in February through March for all districts, while chemical control should be initiated in April in Limulunga district and delayed to May for the rest of the districts. The study has also identified other hotspots of PMD, in addition to Limulunga, that should be priority targets for disease management to maximize the use of limited resources.

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Interactive Effect of Nitrogen Fertilizer and Plant Density on Yield of Nerica 4 Upland Rice using Dibbling Method

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***Abstract*— *The experiment was conducted at the JICA-Tsukuba International Center Experiment Field during the April –October 2015 cropping season. The objective of this experiment was to evaluate the influence of Nitrogen fertilizer and plant density on growth and yield of NERICA 4 upland rice.* **In this** *study, a split-plot experimental design was used with three replications. The treatments comprised of Nitrogen fertilizer at 0 and 60 Kg N/ha; while planting was done using the dibbling method at spacings of 30cm x 30cm, 30cm x 15cm, and 20cm x 15cm which resulted into plant densities of 11.1 hills/m^2, 22.2 hills/m^2 and 33.3 hills/m^2 respectively. Results showed that Nitrogen application increased tiller number, plant length, leaf area index and SPAD Value at both maximum tillering and heading stages. The analyzed data on yield and yield components at (HSD 5%) showed no significant difference in panicle number/m^2, spikelet number/panicle, percentage of ripened grains, 1000 grains weight and paddy yield between plant densities at both 0, and 60 kg N/ha. However, plant density of 22.2 hills/m^2 resulted in the highest paddy yield of 2.79 t/ha and 3.77 t/ha at both Nitrogen levels 0, and 60 kg N/ha respectively. Plant density S_2 (22.2 hills/m^2) was the optimum for NERICA 4 upland rice for increased growth and yield*

***Keywords*— Nitrogen, Fertilizer, Plant density, Yield.**

I. INTRODUCTION

Rice (*Oryza sp.*) is increasingly becoming a crop of high demand both in Uganda and Zambia. Due to the rise in consumption levels of the commodity in these countries, there have been great efforts by respective governments through responsible ministries to double the efforts in its production. Despite these efforts, however, there have been many challenges that have kept production levels at its lowest. In Zambia, lack of knowledge among farmers and extension staff on rice production techniques and related agronomic practices such as planting methods, seed production, variety selection, fertilizer application and timing, have brought about the poor yields (www.iapri.org.zm, 2016). The fertility of Uganda soils is on the whole, declining. This is due to poor farming practices characterized by low inputs use, among other factors, and a generally poor farmers' response to soil conservation practices. The decline in native fertility is worsened by soil erosion and deforestation on top of low input continuous cultivation (Semalulu and Butegwa, 2000).

Plant response to Nitrogen is generally lower in dry than in wet soils because water deficits prevent plants from making full use of Nitrogen. Almost all upland rice soils have low Nitrogen (Aragon and De Datta, 1982; Mahapatra and Shrivastava, 1983). Local production is low and efforts to increase production are hindered by high input costs, and low yields, especially in the uplands. Cultural practices especially plant density (spacing) influence upland rice response to Nitrogen fertilizer (Chaturvedi, 2005). He further advises that judicial use of fertilisers can incredibly increase both the quantity and quality of harvested rice. The objective of this experiment was to evaluate the influence of Nitrogen fertilizer and plant density on growth and yield of NERICA 4 upland rice

II. MATERIALS AND METHODS

The experiment was conducted at the JICA Tsukuba International Center Experiment field, during the April-October 2016 cropping season. The upland rice variety NERICA 4 was used and sown at a seed rate of 30-40 kg ha^{-1}. The experiment was laid out in split plot design with three replications. The plot size was 3.6 m x 3.0m. Nitrogen fertilizer was applied at two levels of 0, and 60 Kg N ha^{-1} ; and plant spacing of 30cm x 30 cm, 30cm x 15 cm, and 20cm x 15 cm, planted using the dibble method with 6 seeds per hill. Data was collected on heading dates, plant height, tiller number, Leaf area index, panicle number, panicle length and 1000 grain weight.

TREATMENTS

N_0 and N_{60} denote fertilizer levels 0, and 60 Kg N ha^{-1} respectively.

S_1: Spacing of 30 x 30 cm (11.1 hills/m^2)

S_2: Spacing of 30 x 15 cm (22.2 hills/m^2)

S_3: Spacing of 20 x 15 cm (33.3 hills/m^2)

The experimental soil was slightly acidic, with optimum amount of nitrogen and carbon to nitrogen ratio.

Table 1Chemical property of experimental soils.

Properties	Values
pH (H_2O)	5.6
Total Carbon (%)	3.47
Total Nitrogen (%)	0.29
C/N	11.96
Available P_2O_5 (mgkg^{-1})	73.59

Meteorological data at JICA-Tsukuba 2016

Fig. 1 below shows the weather condition of rice during the experiment. The weather pattern indicated that month of July and August experienced below normal rainfall and extreme temperatures.

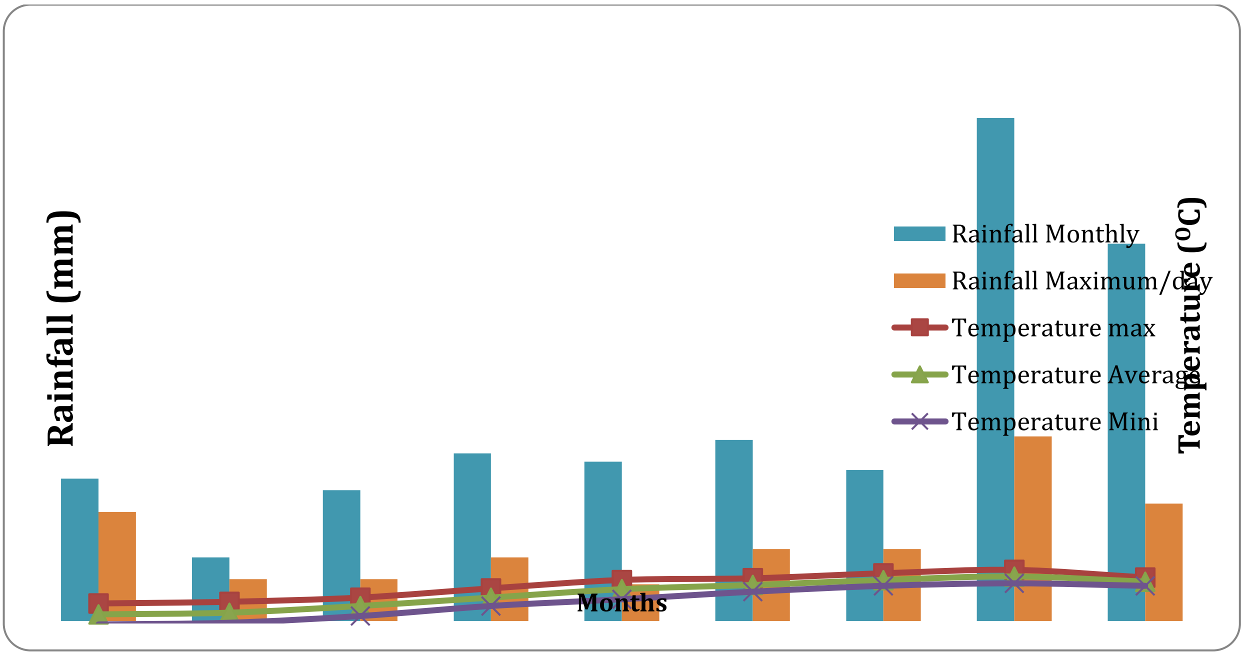

Fig. 1 Temperature and precipitation of the experimental season and normal year (Source: Weather report of the Metrological Agency).

Seed Preparation and sowing

Seeds were selected using 1.06 specific gravity salt solutions, disinfected by Benlate-T (Thiaruam 20%, Benomyl 20%) as fungicides for 24 hours, then dry for 24 hours. After drying seeds were coated with KIHIGENT (TMTD80%) as bird repellent. Seed rate of 30-40kg/ha was used.

Weed Control, GO-GO SAN the herbicides which contains 30% of Pendimethalin was applied 1 day after sowing and 2 times manual weeding was done.

Fertilizer Application

Fertilizers were applied as basal at sowing time and as top dressing about 20 Days before heading (DBH) as shown in Table 2.

Table 2 Fertilizer application regime.

Nutrient Type	Basal Dressing (Kg/ha)	Top Dressing (Kg/ha)	Total (Kg/ha)
1. N_0	0	0	0
2. Nitrogen	40	20	60
3. P (P_2O_5)	100	-	100
4. K (K_2O)	80	20	100

NOTE: N; Ammonium Sulphate (21% N), P_2O_5; Super phosphate (17.5% P); K_2O, Potassium Chloride (61% K).

Agronomic Observation

Agronomic parameters such as seedling establishment rate, SPAD value (chlorophyll content), dry weight/m^2, plant length, tiller number/m^2, leaf area index, culm length, panicle length and heading dates were recorded and evaluated. The panicle number/m^2, spikelet per panicle, percentage of ripened grains and 1000 grain weights were investigated as yield components at maturity stage. For data collection, 100cm rows were marked.

Data Analysis

Excel 2007 was used for data arrangement and the statistical analyses (ANOVA) of data were conducted using Statistics. 9. Mean separation was made according to HSD significance difference with Turkeys test at 5% significant levels.

III. RESULTS

YIELD AND YIELD COMPONENTS

The analyzed data on yield and yield components at (HSD 5%) showed no significant difference in panicle number/m^2, spikelet number/panicle, percentage of ripened grains, 1000 grains weight and paddy yield between plant densities at both 0 Kg and 60 kg of Nitrogen per hectare. However, plant density S2 (22.2 hills/m^2) showed the highest paddy yield at both Nitrogen levels of N_0 2.79 t/ha and N_{60} 3.77 t/ha respectively.

Table 3 Yield and yield components

Nitrogen Level, NL (Kg/ha)	Plant Density, PD (hills/m^2)	Panicle Number/m^2	Spikelet Number/panicle	Percentage of Ripened Grains (%)	1000 grains Weight (g)	Paddy Yield (t/ha)
N_0	S_1	140.30 a	89.10 a	63.85 a	27.42 a	2.34 a
	S_2	150.93 a	134.57 a	54.70 a	27.91 a	2.79 a
	S_3	172.90 a	76.30 a	64.05 a	26.76 a	2.18 a
N_{60}	S_1	129.63 a	121.70 a	59.00 a	28.11 a	2.69 a
	S_2	192.60 a	128.23 a	55.00 a	28.77 a	3.77 a
	S_3	183.33 a	73.37 a	78.30 a	27.95 a	2.95 a
NL X PD		ns	ns	ns	ns	ns

In each column, common letters indicate non-significant difference by Tukeys Honestly Significant Difference (HSD) at 5% level; ns not significant

CORRELATIONS

The analyzed results showed a high positive correlation (r=0.73) between number of panicles/m^2 and paddy yield as shown in Figure 2.

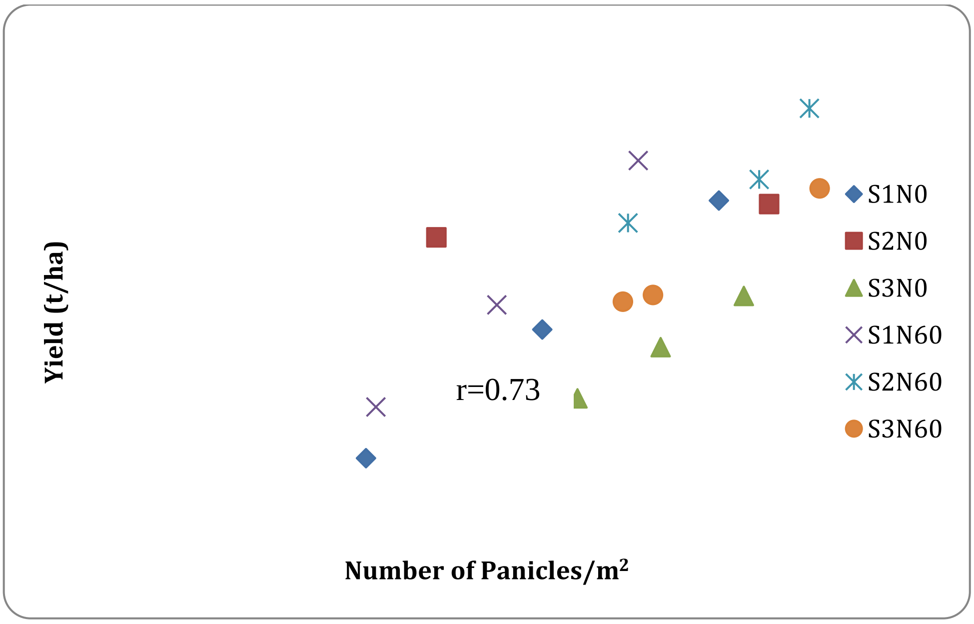

Fig. 2 Correlation between Number of Panicles/m^2 and Paddy Yield.

From the results, there was correlation (r=0.44) between number of Spikelets/panicle and paddy yield as shown in Figure 3.

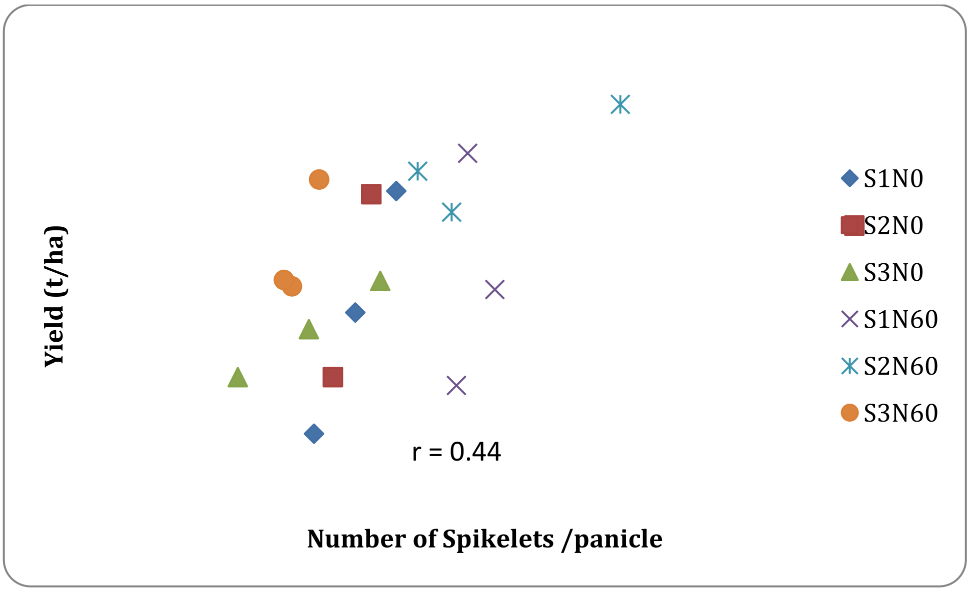

Fig. 3 Correlation between Number of Spikelets/ Panicles and Paddy Yield.

Nitrogen accumulation

The results showed no significant differences in Nitrogen accumulation between Nitrogen levels and planting densities throughout the three growth stages. At both maximum tillering and heading stages, planting density S_3 showed the highest Nitrogen accumulation at Nitrogen level 0 kg/ha. But at maturity stage S_2 planting density S_2 (22.2 hills/m^2) had the highest amount of accumulated Nitrogen.

However, at Nitrogen 60 kg/ha throughout all the three growth stages, S_2 (22.2 hills/m^2) showed the highest nitrogen accumulation as shown in the Table 2 below.

Table 4 Nitrogen Accumulation.

Applied Nitrogen (Kg/ha)	Plant Density (hills/m²)	Nitrogen Accummulation (%)						
		Maximum tillering	Heading Stage			Maturity Stage		
			Culm	Panicle	Total	Culm	Panicle	Total
N_0	S_1	1.60 a	1.77 a	0.83 a	2.6 a	2.63 a	2.63 a	5.3 a
	S_2	1.93 a	2.98 a	1.28 a	4.3 a	3.24 a	3.24 a	6.5 a
	S_3	2.05 a	3.18 a	1.59 a	4.8 a	2.88 a	2.88 a	5.8 a
N_{60}	S_1	1.90 a	2.99 a	1.92 a	4.9 a	3.89 a	3.89 a	7.8 a
	S_2	3.77 a	5.19 a	3.86 a	9.1 a	5.69 a	5.69 a	11.4 a
	S_3	2.83 a	2.88 a	4.48 a	7.4 a	3.01 a	3.01 a	6.0 a

Means in a column with the same letter are not significantly different at 5% level by Tukey's HSD

TILLER NUMBER

Fig. 6 and Fig. 7 show no significant differences between Nitrogen levels and plant densities on tiller number for S_1 (11.1 hills/m²) and S_3 (33.3 hills/m²) at both Maximum tillering and heading stages. Nitrogen levels had no statistical influence on tiller number. However, at both stages of growth, Nitrogen levels increased tiller number at planting densities S_2 (22.2 hills/m²).

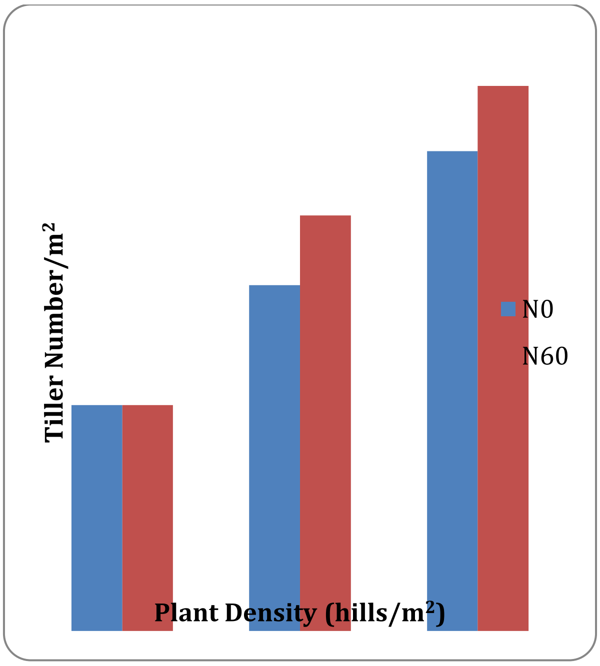

Fig. 6 Effect of Nitrogen fertilizer and plant density on plant length at maximum tillering stage.

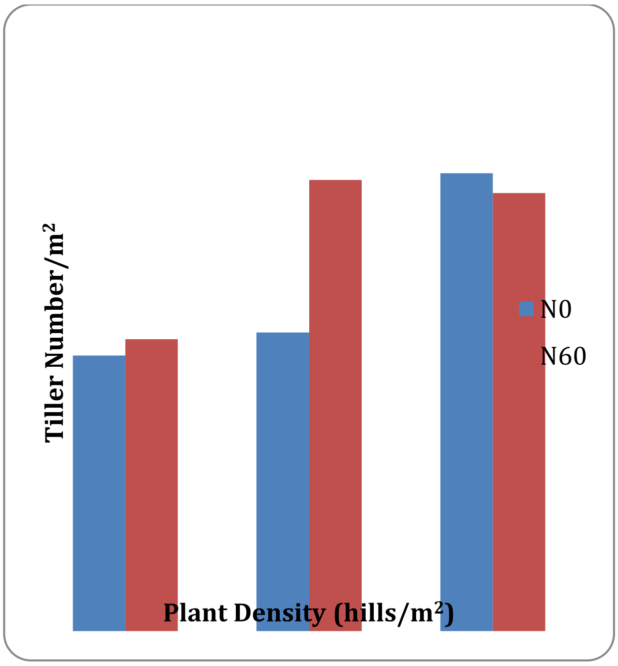

Fig. 7 Effect of Nitrogen fertilizer and plant density on plant length at headingstage.

There were no significant differences between Nitrogen levels and Plant densities on plant length at both maximum tillering (65 DAS) and heading stage (93 DAS). The study showed that Nitrogen levels influenced plant length at both planting densities with S^2 (22.2 hills/m²) showing the longest plant length (Fig. 8, Fig. 9).

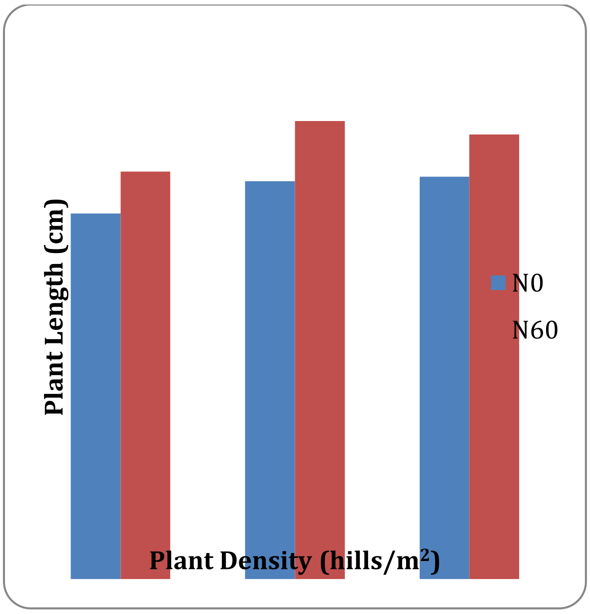

Fig. 8 Effect of Nitrogen fertilizer and plant density on plant length at maximum tillering stage.

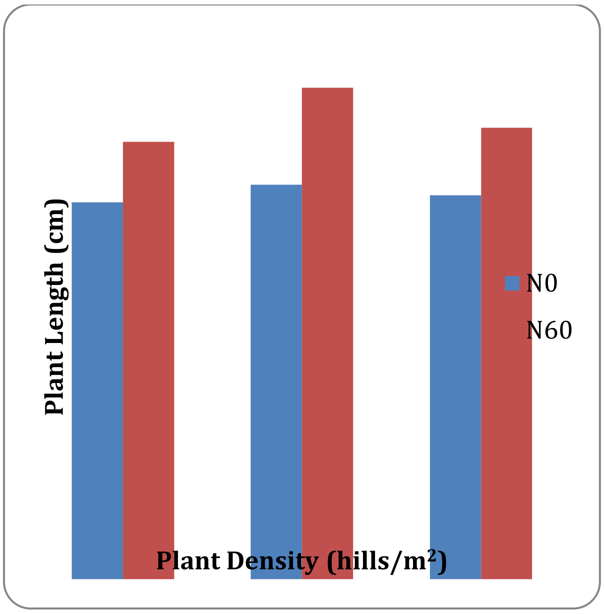

Fig. 9 Effect of Nitrogen fertilizer and plant density on plant length at heading stage.

SPAD VALUE

The observed results showed that S1 and S_2 had the highest SPAD values at maximum tillering (65 DAS), showing distinct influence of Nitrogen and plant density interaction on SPAD value. But S_3 did not show any responsiveness to Nitrogen levels and planting densities on SPAD value. However, at heading stage (93 DAS), S_3 (33.3 hills/m^2) showed the highest SPAD value followed by S_2 and the least was S_1. There were significant increases in SPAD Value with N application. SPAD Value increased with Nitrogen application (Fig. 10, Fig. 11).

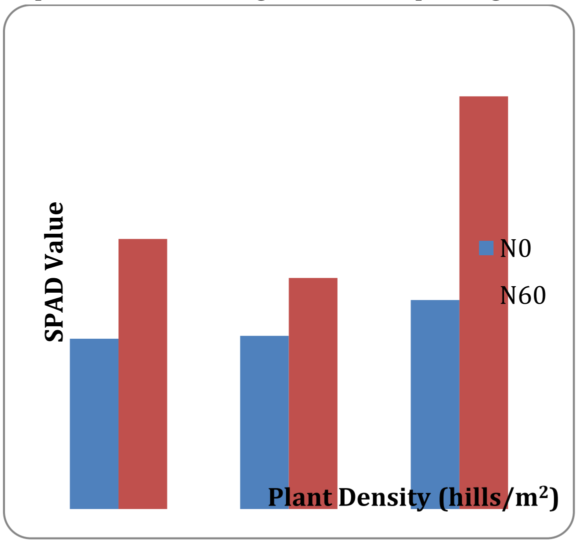

Fig. 10 Nitrogen level and plant density on SPAD value.

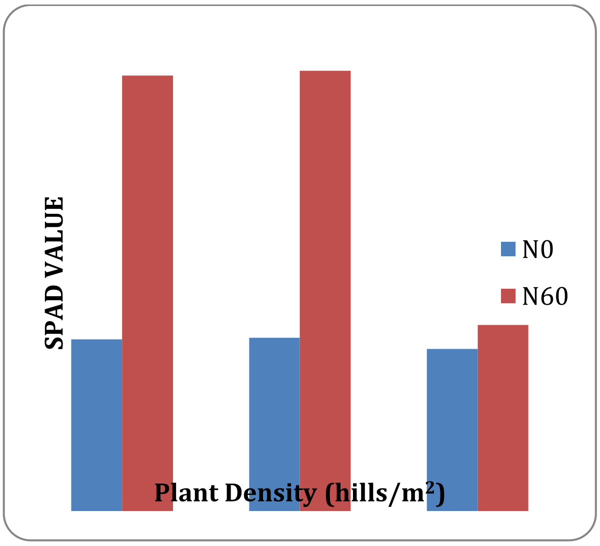

Fig. 11 Nitrogen level and plant density on SPAD value.

LEAF AREA INDEX

From the ANOVA, there were no significant differences between Nitrogen levels and Plant density on leaf area index at both maximum tillering (65 DAS) and heading stages (93 DAS). However, in both stages of growth, S_2 showed the highest LAI at N_{60} (Fig. 12, Fig. 13).

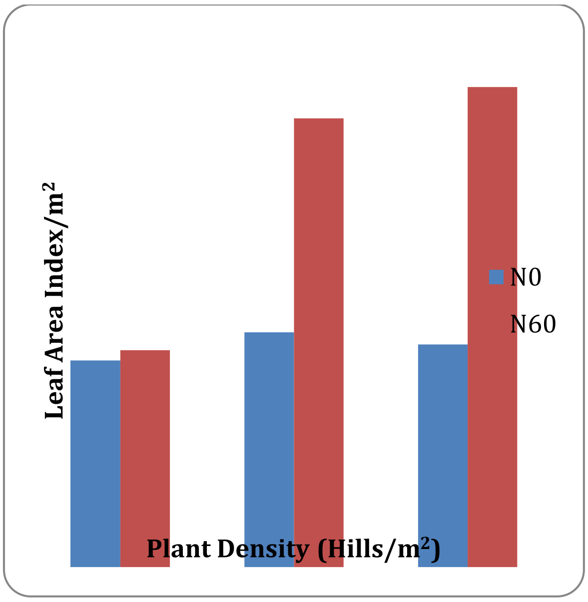

Fig. 12 Nitrogen and plant density on LAI.

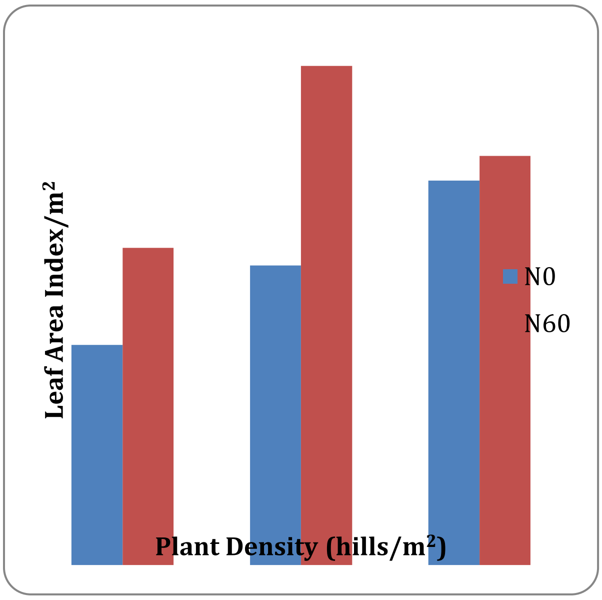

Fig. 13 Nitrogen and plant density on LAI.

IV. DISCUSSION

From the ANOVA, S_2 (22.2 hills/m^2) showed the highest paddy yield for both N_{60} (3.77 t/ha) and N_0 (2.79 t/ha). This can be attributed to the Panicle number/m^2 and spikelet number/panicle which was highest among the three planting densities at both Nitrogen levels. However, this was not the case for S_1 (11.1 hills/m^2) and S_3 (33.3 hills/m^2) which showed lowest paddy yields at both levels of Nitrogen. This is because S_2 was highly responsive to Nitrogen, resulting in less competition for nutrients and light amongst plants per unit area. These findings were confirmed by Fageria and Baligar (2001) who reported that grain yield in rice is a function of panicles per unit area, number of spikelets per panicle, 1000-grain weight and spikelet sterility. Further studies by Kamara and others (2001) reported similar findings that number of grains per panicle increased with increment in nitrogen rates. This was confirmed by the strong positive correlation (r=0.73) between number of panicles/m^2 and paddy yield. And a positive correlation (r=0.44) between number of spikelets per panicle and paddy yield. It is therefore, important to understand the management practices that influence yield components and consequently grain yield

No significant differences were observed in dry matter and Nitrogen Content accumulation in all the three stages of growth between nitrogen levels and Plant densities. But S_2 (22.2 hills/m^2) showed the highest dry matter and Nitrogen content at N_{60}. The dry matter content was higher in treatments with Nitrogen, the highest being at S_2 (22.2 hills/m^2) and N_{60} in all growth stages. This was due to less competition for nutrients and high responsiveness to Nitrogen fertilizer application.

It was further observed that Nitrogen levels and plant densities increased tiller number, plant length and leaf area index at all the three stages of growth. Amanullah *et al.*, (2008) reported similar findings that the increase in leaf area index increases light interception and hence photosynthetic activities are higher. This is due to high responsiveness to Nitrogen.

ANOVA results showed that SPAD value increased with fertilizer application in both S_1 (11.1 hills/m^2) and S_2 (22.2 hills/m^2) due to high photosynthetic activities and access to light. This caused a decrease in chlorophyll accumulation due to shading effect and competition for nutrients was low in S_3 (33.3 hills/m^2) which resulted in low SPAD Value

In conclusion, Nitrogen application increased tiller number, plant length, leaf area index and SPAD Value at both maximum tillering and heading stages. Plant density S_2 (22.2 hills/m^2) is the optimum for NERICA 4 upland rice for increased growth and yield. The recommendation is that a similar study must be conducted in both Uganda and Zambia to ascertain the optimum nitrogen level and plant density combination.

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Identification of therapeutic potential phytochemicals against the SARS CoV-2 spike protein of omicron variant by Molecular docking techniques

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***Abstract*— *Repeated mutations in the SARS CoV-2 have resulted in the emergence of various life-threatening variants including omicron, triggering a systemic crisis in global human health. Due to the unavailability of any specific antiviral treatment, the effectiveness of the currently available drugs or vaccines against this variant remains doubtful. Phytochemicals from their known medicinal plants were selected with the antiviral, antifungal, anti-inflammatory, and antioxidant properties and screened to study their therapeutic potential and druglikeliness properties. A library **of 15** phytochemicals from various medicinal plants was constructed and their binding energies were calculated by docking them against the spike protein of the SARS CoV-2 omicron variant using the AutoDock software. Subsequently, the pharmacokinetic properties like ADME and drug likeliness properties were also calculated. Hydroxychloroquine (HCQ), an antimalarial drug was chosen as the standard in docking analysis, displaying the binding energy of -5.22 kcal/mol whereas, Glycyrrhetic acid showed the highest binding energy with the value of -9.02 kcal/mol. Besides, all these 15 phytochemicals showed higher binding energy (≤ -7.00 kcal/mol) with the better pharmacokinetic properties leading them a viable drug candidate to treat the infection caused by omicron variant.*

***Keywords*— Drug likeliness, Insilico studies, Omicron, Phytochemicals, SARS CoV-2.**

I. INTRODUCTION

The emergence and persistence of the coronavirus pandemic that was first identified in Wuhan, China in late December 2019, is still ongoing with a significant rise in the rate of mortality and morbidity.

According to the World Health Organization (WHO) dashboard report, till 28th February 2022, 434,154,739 confirmed cases have been reported globally, including 5,944,342 deaths [1], and an increase in the number of cases has been observed day by day, thus raising the great public health concerns worldwide.

COVID-19 is an infectious disease caused by SARS-CoV-2 (Severe Acute Respiratory Syndrome Coronavirus-2). Genomically, this virus is related to two known CoV-strains, namely the Middle East Respiratory Syndrome Coronavirus (MERS-CoV) emerged in 2012, and the Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) in late 2003 [2].

The disease is characterized by symptoms such as fever, dry cough, headache, sore throat, fatigue, diarrhoea, and dyspnea followed by acute respiratory distress syndrome [3] and is predominantly known to spread from person-to–person [4]. In addition to this, the virus is also known to infect the upper and lower respiratory tract, liver, kidney, gut, heart, and nervous system and eventually lead to multi-organ damage [5]. Immunocompromised people with obesity, hypertension, diabetes, and cardiovascular disorders are found to be more susceptible[6].

SARS CoV-2 is an enveloped, positive-sense single-stranded RNA virus with a genome size of 26-32 kb [7][8], and is known to be transmitted between animals and humans [9], belonging to the family Coronaviridae and genus Coronavirus [10].

The viral genome consists of 14 functional open reading frames (ORFs) including two noncoding regions at both ends followed by the multiple regions that encode for 29 proteins including structural, nonstructural proteins (NSP), and accessory proteins. ORF 1a and ORF 1b encode 16 NSPs (nsp1-nsp16) such as Helicase (nsp13), RNA-dependent RNA polymerase (nsp12), Papain-like protease (nsp3), main protease (nsp5) also known as 3C-like protease (3CLpro), and 2O methyltransferase (nsp16), which are likely to be involved in transcription, replication, and pathogenesis and play a vital role in the life cycle of the pathogen. Whereas structural proteins include, Spike surface glycoprotein (S), a small envelope protein (E), membrane protein (M), and nucleocapsid protein (N), which are essential for viral assembly and the production of a structurally whole viral particle. And the remaining 9 are the accessory proteins that provide a selective advantage in the infected host cell [11] [12].

The S protein plays an integral role in viral attachment through the recognition of host cell receptors and promotes the fusion of the viral and host cell membranes, to facilitate the viral entry into the host cells. This protein is composed of two functional subunits: an amino (N)-terminal S1 subunit and a carboxyl (C)-terminal S2 subunit with a single transmembrane region anchor. The S1 subunit consists of an N-terminal domain (NTD) and a receptor-binding domain (RBD), along with two C-terminal domains. Whereas the S2 subunit consists of a fusion peptide (FP), heptad repeat 1 (HR1), central helix (CH) region, connector domain (CD), heptad repeat 2 (HR2), transmembrane (TM) region, and the cytoplasmic tail (CT). The cleavage site at the border between the S1 and S2 subunits is called S1/S2 protease cleavage site [13].

In the native state, the S protein exists as an inactive precursor. During the infection, the S protein gets activated by the TM protease serine 2 (TMPRSS2), a type 2 TM serine protease located on the host cell surface, by cleaving the S protein into S1 and S2 subunits, that are required for activating the membrane fusion domain after the entry of the virus into the host cell [14]. Later, the S1 subunit is shed and the S2 subunit undergoes large conformational change compared to its prefusion state that mediates the membrane fusion process, thus, inserting the fusion peptide into the host cell membrane, followed by the release of viral RNA into the host cell [13], where the polyproteins are translated from the RNA genome. Protein cleavage and the assembly of the replicase–transcriptase complex, make the replication and transcription process of the viral RNA genome occur. The replicated viral RNA and the structural proteins that are newly synthesized are assembled and packaged in the host cell before the release of viral particles [14].

Since, the S protein plays an active role in the recognition, attachment, and fusion of the viral particle into the host cell, thus designing an effective antiviral drug or inhibitor by targeting this protein can be the best solution to treat the infection.

SARS CoV-2 virus, which was previously identified in Wuhan, China in late 2019, has undergone multiple significant mutations resulting in the new variants including Alpha, Beta, Gamma, Delta, Omicron, and so on.

Out of these variants, Omicron is one of the most rapidly spreading SARS CoV-2 variants across the world and has been classified as a “variant of concern (VOC)” by the World Health Organization (WHO) [15].

The first case of the omicron variant (B.1.1.529.1), was reported in South Africa on 23 November 2021 [16] and as of 14 December 2021, it had spread to more than 76 countries [17]. This variant is known to be the highly mutated strain compared to the other VOCs (Alpha, Beta, Gamma, and Delta), with 50 mutations accumulated throughout the genome. Out of which, at least 32 mutations are found in the spike protein, which is twice as many as the Delta variant. These mutations affect the biological characteristics of the omicron variant, by increasing the transmissibility, causing immune escape, and enhancing the virulence. One of the Insilico studies showed that the infectivity of the Omicron variant might be more than 10-fold higher than that of the original virus (Wuhan) [18][19].

The emergence of this variant has posed an increased risk to the global public health, concerning the effectiveness of the currently available drugs or vaccines. At present, most clinical studies are in progress, to study the effect of antiviral drugs, corticosteroids, antimalarials, and monoclonal antibodies against this variant.

Currently, only a few repurposed FDA-approved drugs are being used to treat the infection including Remdesivir, Lopinavir/Ritonavir, Oseltamivir, Favipiravir, Rifampicin, Letermovir, Azithromycin, Cholorquine, and Hydroxychloroquine [20]. Corticosteroids including dexamethasone [21]and methylprednisolone [22], are also being used to treat the infection.

Along with this, scientists worldwide are studying the effectiveness of these drugs alone or with a combination of other antiviral drugs. However, few of the drugs showed certain side effects like cardiotoxicity, hematologic toxicity, hepatotoxicity, and nephrotoxicity.

E.g., Chloroquine and Hydroxychloroquine which were found promising earlier have resulted in neuromyopathy, cardiomyopathy, and retinopathy and was found to decrease in-hospital survival and increased the frequency of ventricular arrhythmias in hospitalized patients [23].

Serum neutralization assay against the omicron variant showed that the neutralization activity of the Pfizer/BNT162b2 mRNA vaccine declined the serum activity by 41.4 fold compared to the original strain (Wuhan) [18][24]. In addition to this, an in vitro study of imdevimab and casirivimab was found to be resistant to authentic omicron variant, which was effectively used to prevent Delta infection [25]. Thus, with the decrease in the vaccine efficacy and unavailability of any specific antiviral drug candidate against the omicron variant, the world is still battling to overcome the Pandemic.

India is home to more than 8000 species of medicinal plants [26]. These plants have been known for their immense potential and tremendous properties against various infectious diseases and health-related complications from ancient times. However, these plants are a rich source of chemically active compounds including alkaloids, flavonoids, terpenoids, polyphenols, tannins, saponins, etc. possessing a variety of biological deliberations, including antioxidant, antibacterial, antifungal, anti-inflammatory, and antiviral properties [27].

At present, plant-based phytochemicals are attracting the focus of modern world healthcare researches, for developing a vaccine or treatment against the infection, since they are known to be less toxic with minimal side effects. It is reported that approximately 70–80% of mainstream medicines originated from natural products [28]. In addition to these, plant-based phytochemicals offer attractive, effective, and holistic drug action against pathogens. Thus, extracting the plant-based compounds and herbal treatments from these plants can be a cure and a solution against omicron infection.

In this regard, Insilico studies were carried out to identify the effect of the phytochemicals that can play a role as an inhibitor by studying their binding affinity towards the spike protein. In the modern world, Insilico studies are gaining much attention worldwide for their advanced strategies and effective techniques for identifying putative drug candidates that can be further led to in vitro and in vivo assessments. These methods basically provide the most accurate results, require shorter time duration, are cost-effective, and fall in the dry lab category, thus being widely used in understanding and predicting the drug ability in the early stages of drug discovery [29].

The current study is aimed to explore the drugs with a therapeutic potential action of the phytochemicals as an effective inhibitor against the spike protein of omicron variant, using Computational pharmacology and virtual screening techniques. A library of 15 phytochemicals was prepared from various medicinal plants, based on their therapeutic potential against infectious diseases. The binding energy of all these phytochemicals was calculated and compared with that of HCQ (an antimalarial drug) which was chosen as the standard reference drug for comparison. Additionally, ADME analysis and drug likeliness studies were also conducted for understanding the safety and efficacy of the phytochemical, which can play a role as the drug candidate for the further development of an effective drug or inhibitor against the infection caused by the omicron variant.

II. MATERIALS & METHODOLOGY

2.1 Preparation of receptor:

The cryo-electron microscopic 3-Dimensional structure of SARS CoV-2 Omicron variant spike protein was downloaded from RCSB-Protein Data Bank with PDB ID: 7T9J [30] and was visualized by using Chimera software 1.14 [31]. In this study, Spike glycoprotein was chosen as the target site. Since, it plays a crucial role in its attachment, fusion, and viral entry into the host cell. The spike protein of the omicron variant consists of 3 chains, Chain A, B, and C as shown in Fig. 1. Out of which, Chain C (represented in Fig. 2) was selected for the docking process, eventually energy minimization and protein optimization was carried out by using the Swiss PDB Viewer [32].

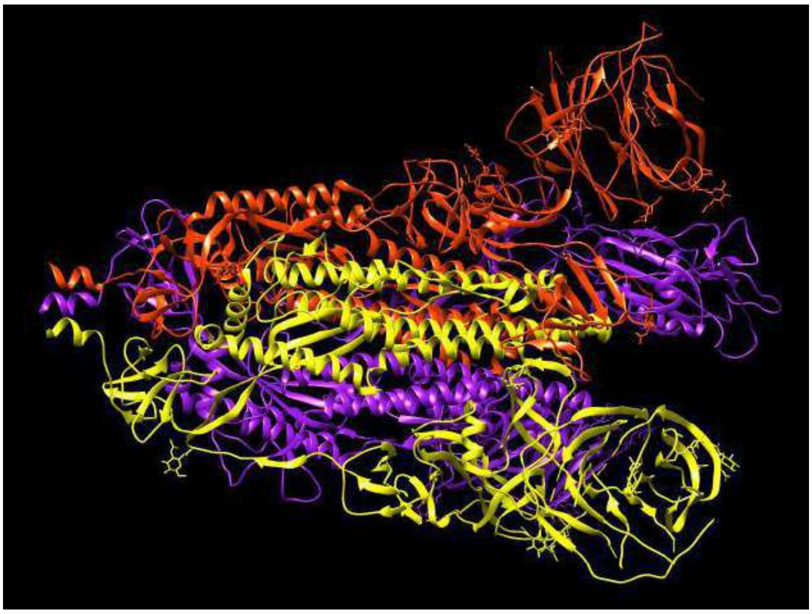


Fig.1: 3D structure of SARS CoV-2 omicron variant spike protein (PDB ID: 7T9J), Chain A (orange-red colored), Chain B (yellow colored), & Chain C (purple colored)

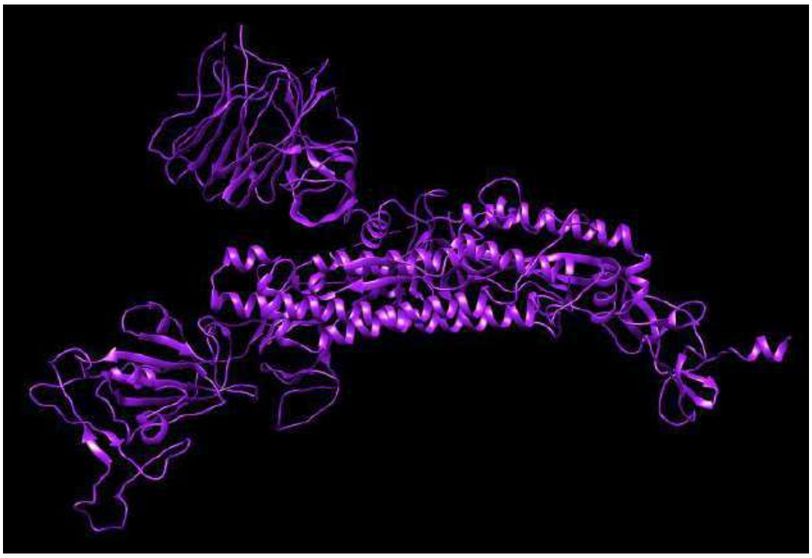
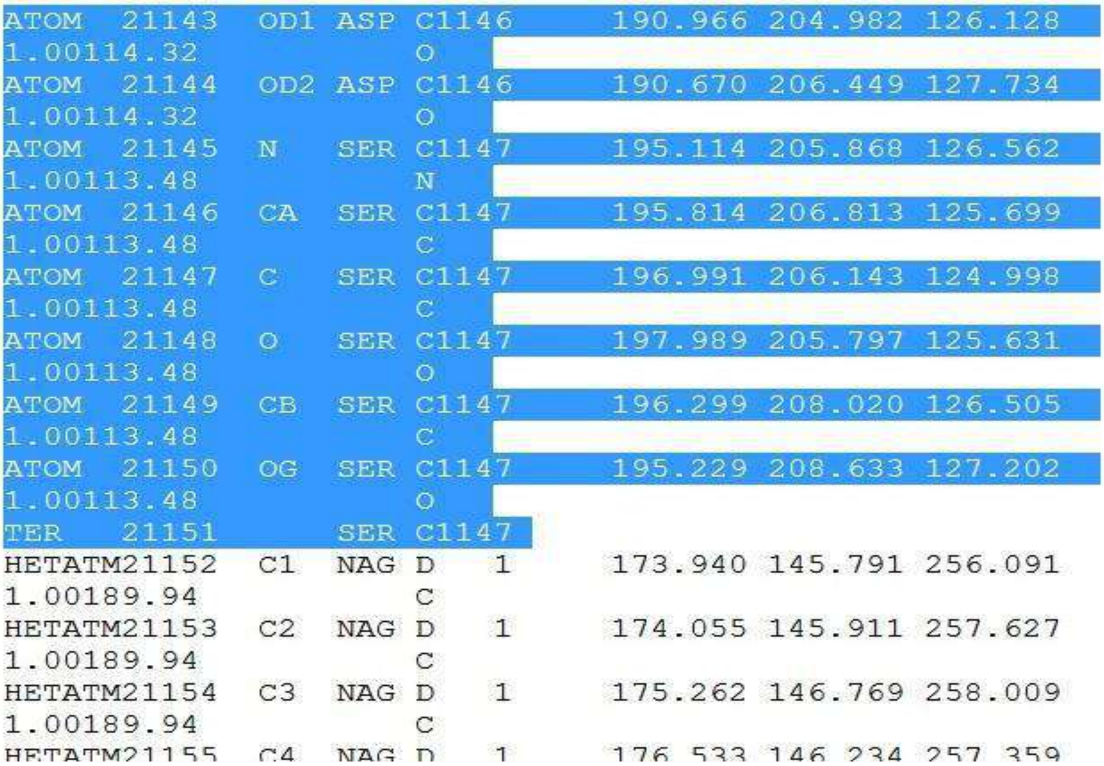


Fig.2: 3D structure of Chain C of SARS CoV-2 omicron variant spike protein

2.2 Protein optimization and energy minimization:

This step is carried out to remove the additional molecules and the extra spaces from the pdb protein files, by arranging the protein coordinates into the proper order. The pdb structure of the protein downloaded from the RCSB-PDB database was opened in WordPad and the individual chain (Chain C) of the protein was retrieved (represented in Fig. 3) and saved in 'all files' format.



```
ATOM  21143  OD1 ASP C1146      190.966 204.982 126.128
1.00114.32           O
ATOM  21144  OD2 ASP C1146      190.670 206.449 127.734
1.00114.32           O
ATOM  21145  N   SER C1147      195.114 205.868 126.562
1.00113.48           N
ATOM  21146  CA  SER C1147      195.814 206.813 125.699
1.00113.48           C
ATOM  21147  C   SER C1147      196.991 206.143 124.998
1.00113.48           C
ATOM  21148  O   SER C1147      197.989 205.797 125.631
1.00113.48           O
ATOM  21149  CB  SER C1147      196.299 208.020 126.505
1.00113.48           C
ATOM  21150  OG  SER C1147      195.229 208.633 127.202
1.00113.48           O
TER   21151      SER C1147
HETATM21152  C1  NAG D   1      173.940 145.791 256.091
1.00189.94           C
HETATM21153  C2  NAG D   1      174.055 145.911 257.627
1.00189.94           C
HETATM21154  C3  NAG D   1      175.262 146.769 258.009
1.00189.94           C
HETATM21155  C4  NAG D   1      176.533 146.234 257.359
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Fig.3: Protein Optimization using WordPad

Energy minimization of the protein chain was carried out by using the Swiss PDB Viewer tool (v4.1.0), followed by removing the additional swiss coordinates that were added, by using WordPad.

2.3 Ligand preparation:

A library of 15 phytochemicals from numerous medicinal plants was constructed, based on their therapeutic properties. The 3-Dimensional structures of all these phytochemicals including HCQ (PubChem Id: 3652), an antimalarial drug chosen as a standard reference drug were downloaded from the PubChem database in structure-data file (sdf) format. Later, the structure-data file (.sdf) format of these phytochemicals was converted into the pdb format by using Open Babel software 2.4.1 [33] and was visualized in Chimera software 1.14. The pictorial representation of the 3D structure of Glycyrrhetic acid visualized by using Chimera software is represented in figure 4.

Fig.4: 3D structure of Glycyrrhetic acid.

And Table 1 consists of the name list of all the 15 selected phytochemicals with their PubChem ID's and the scientific name of the medicinal plant from which it is obtained followed by their therapeutic properties.

Table 1: List of phytochemicals isolated from few selected medicinal plants.

Botanical name of the plant	**Phytochemical**	**PubChem Id**	**Therapeutic properties**
Elodea canadensis	β- Sitosterol	222284	Antioxidant [34][35], anti- inflammatory drug [36] and antiviral [37].
Syzygium claviflorum	Betulinic acid	64971	Anti-HIV [38], antimalarial [39], anti-inflammatory, antioxidant and anticancer [40].
Desmodium gangeticum	Desmodin	13338925	Antioxidant [41] and used in the treatment of Alzheimer's disease [42].
Momordica foetida	Foetidin	15945065	Anti-diabetic [43].
Glycyrrhiza glabra	Glycyrrhetic acid	10114	Antiviral [44], immunomodulatory, anti-inflammatory and hepatoprotective [45].
Ferula assa-foetida	Kamolonol	46883037	Antibacterial [46] and cardioprotective property [47].
Marrubium globosum	Marrubiin	73401	Antioxidant, anti- diabetic antinociceptive, antigenotoxic, antioedematogenic, antispasmodic, analgesic, gastroprotective, immunomodulating, cardioprotective and vasorelaxant properties [48].
Salvia tomentosa	Maslinic acid	73659	Anti-inflammatory [49], anticancer [50], anti diabetic [51], antioxidative, cardioprotective [52]and neuroprotective properties [53].
Ophiopogon japonicas	Oleanolic acid	10494	Antioxidative, antiviral, anti-inflammatory, immunomodulatory and cardioprotective properties [45][54].
Prunella vulgaris	Oleanane	9548717	Anticancer, antimicrobial, anti- inflammatory, cytotoxic and hepatoprotective [55].
Gladiolus italicus	Ursolic acid	64945	Antiviral [56], antioxidant, anti-inflammatory and anticancer [45].
Withania somnifera	Withaferin A	265237	Anti platelet, anti herpetic, anti-inflammatory, anticancer, antileishmanial and immunosuppressive properties [57].
Withania somnifera	Withanone	21679027	Antiviral [58], anticancer [59] and neuroprotective [60].
Withania somnifera	Withanolide	53477765	Antiviral [61], anti-inflammatory Immunomodulatory and anti-cancer properties [62].
Plumbago zeylanica	Zeylanone	5276618	Antimicrobial and cytotoxic properties [63]

2.4 Prediction of Active Binding site

An extensive literature survey was conducted to predict the active binding sites on the receptor molecule for docking. Few of the reports suggested that the amino acid residues such as R 493, S 496, and R 498 are considered the active binding amino residues present on the spike protein of the omicron variant [30][64][65].

2.5 Molecular Docking of Receptor and Ligand

The probable inhibitory effects of each of the phytochemicals were evaluated and compared with the std. HCQ by the docking method using AutoDock software 1.5.6 [66]. Prior to the docking process, the protein and the ligands were prepared with the addition of polar hydrogen molecule and Gasteiger charges by removing the water molecules and a few torsion adjustments were made specifically for ligands and were saved in pdbqt format.

The dimension of the grid box was selected as 38Å X 44Å X 56Å with the grid values of X - coordinate = 203.458, Y – coordinate = 181.765 and Z – coordinate = 269.208. The size and dimension of the grid box were selected based on the position of active binding sites of the omicron variant, with a total of 100 genetic runs, while the other parameters were set as default. During the docking process, the receptor (spike protein) was kept rigid and the ligand (phytochemical) was flexible.

The binding affinities of these phytochemicals towards the targeted protein were identified and compared with HCQ. The data generated from the docking process showed how well the ligand gets interacted with the protein of interest. And these values were further analyzed by using MGL tools 1.5.6 [66].

Later, the output files obtained from the docking process were used to study the molecular interactions between the protein-ligand complexes including hydrogen bonds by using Ligplot+ 2.2 software [67].

2.6 ADME analysis & Drug likeliness

The sdf structure of the phytochemical compounds downloaded from the PubChem database was fed into the SwissADME server [68] and converted into canonical SMILES format. These SMILES structures were used to study the ADME (Absorption, Distribution, Metabolism, and Excretion) properties of the screened phytochemicals, and the data obtained from this showed that how well a chemical drug (phytochemical) is processed in a living organism.

Some of the important parameters related to the drug likeness properties of the compounds including molecular weight, number of hydrogen bond donors, number of hydrogen bond acceptors, and molar refractivity were computed utilizing Lipinski's rule of five [69].

III. RESULTS

The current study was focused on identifying the specific antiviral drug or inhibitor by targeting the spike glycoprotein of the omicron variant by drug repurposing technique since this protein plays a prime role in its attachment and internalization into the host cells. Thus, identifying a new drug or inhibitor which can inhibit the binding of the spike glycoprotein to the host cell can be a solution against the infection.

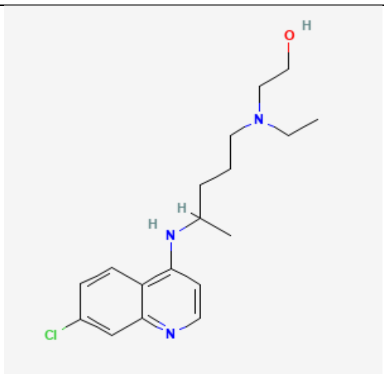
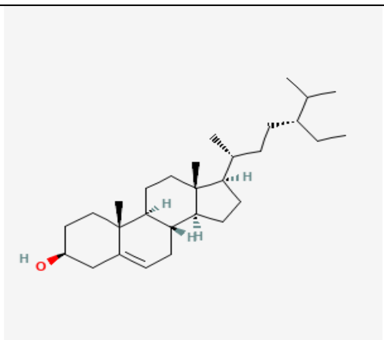
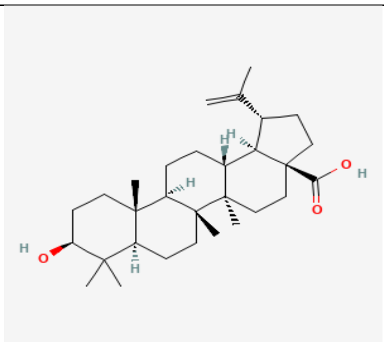
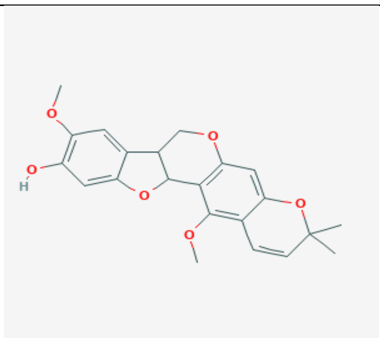
3.1 Virtual Screening and Visualization

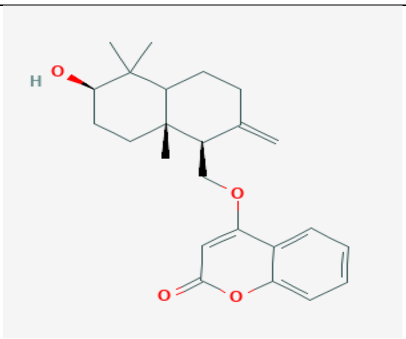
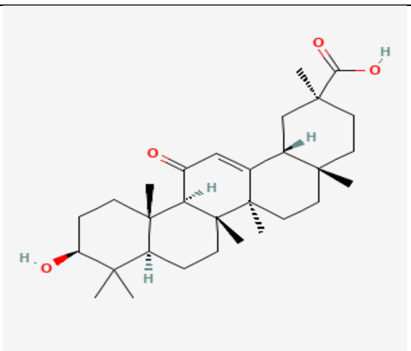
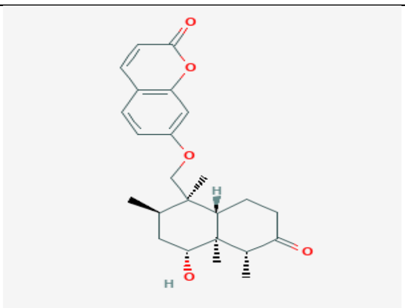
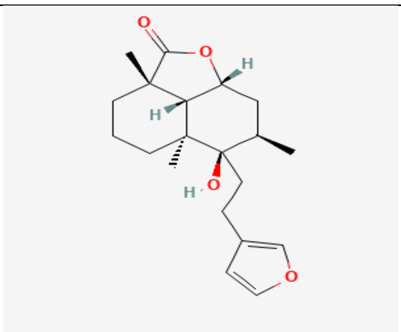
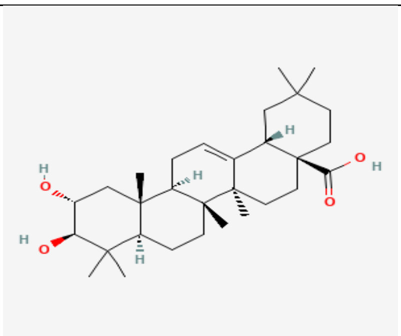
Molecular docking is the most promising tool in the drug discovery process, which is used to predict the binding affinity of the ligand towards the targeted protein molecule when the 3D structure of the target protein is known. It is also used to study the interactions between the protein-ligand complexes at the atomic level and characterize the behavior of ligands in the binding site of target proteins as well as elucidate fundamental biochemical processes [70]. Because of their wide applications, this method is mostly preferred in all pharmaceutical industries. Hence, this method was employed to study the therapeutic potential of the selected phytochemicals including HCQ against the SARS CoV-2 omicron variant based on their binding affinity of the phytochemical (ligand) towards the targeted protein (spike protein).

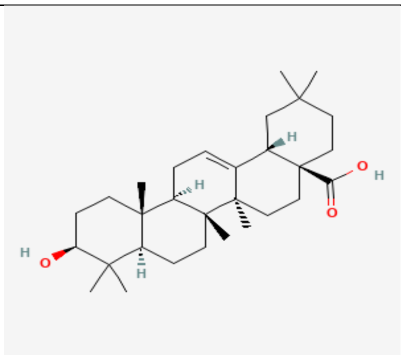
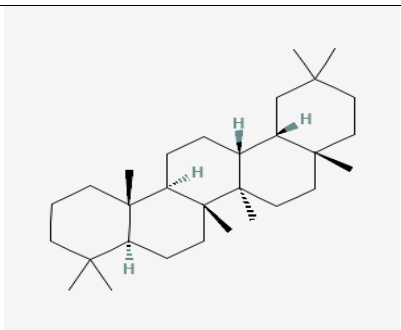
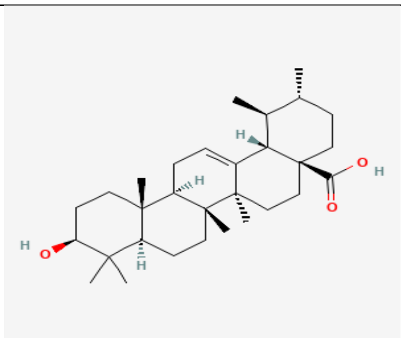
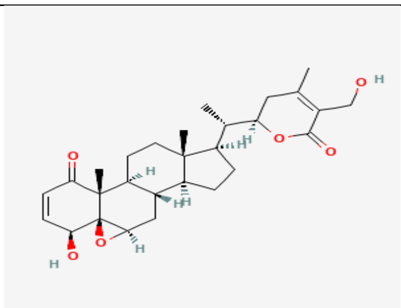
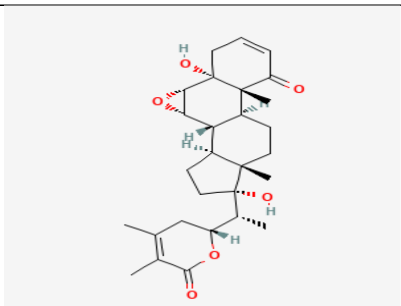
Higher the negative value of the binding energy stronger the binding of the phytochemical towards the targeted protein.

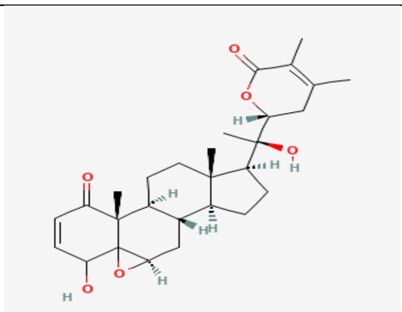
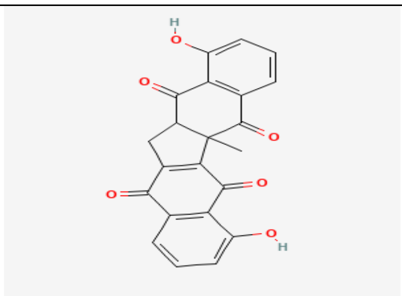
The binding energy of all the 15 phytochemicals against the spike protein of the SARS CoV-2 omicron variant was calculated by AutoDock software 1.5.6. The 2D structure of the selected phytochemicals along with their binding energies and interacting residues are represented in Table 2.

Table 2: The binding energy of the compound towards the spike protein of the omicron variant along with their interacting residues with the grid dimension of 38 X 44 X 56 (Å) and grid value of X = 203.458, Y = 181.765 and Z = 269.208 (The 2D structure of all the ligands represented in the above table are downloaded from PubChem database)

Name of the ligand	**2D-Structure**	**Binding energy (kcal/mol)**	**Interacting amino acid residues**
Hydroxychloroquine		-5.22	Hydrogen bond (H- bond): ARG 403, ARG 493, ARG 498 and HIS 505. Hydrophobic interactions (H-I): TYR 449, TYR 453, SER 494, TYR 495, SER 496, PHE 497 and TYR 501.
β- Sitosterol		-7.45	H- bond: HIS 505. H-I: ARG 403, TYR 453, ARG 493, SER 494, TYR 495, SER 496, PHE 497 and TYR 501.
Betulinic acid		-7.46	H- bond: ARG 493. H-I: ARG 403, TYR 453, SER 494, TYR 495, SER 496, PHE 497 and HIS 505.
Desmodin		-7.07	H- bond: SER 494. H-I: ARG 403, TYR 449, TYR 453, ARG 493, TYR 495, SER 496, PHE 497 and HIS 505.

Foetidin		-7.36	H- bond: ARG 493. H-I: ARG 403, TYR 449, TYR 453, SER 494, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505.
Glycyrrhetic acid		-9.02	H- bond: TYR 453 and SER 494. H-I: ARG 403, ARG 493, TYR 495, SER 496, TYR 501 and HIS 505.
Kamolonol		-7.41	H- bond: TYR 453 and SER 494. H-I: ARG 403, ARG 493, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505.
Marrubiin		-7.09	H- bond: SER 496. H-I: ARG 403, TYR 453, SER 494, TYR 495, PHE 497, TYR 501 and HIS 505.
Maslinic acid		-8.17	H- bond: ARG 493. H-I: ARG 403, TYR 453, SER 494, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505.

Oleanolic acid		-8.68	H- bond: ARG 493. H-I: ARG 403, TYR 453, SER 494, TYR 495, SER 496, TYR 501 and HIS 505.
Oleanane		-7.63	H-I: ARG 403, ARG 493, SER 494, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505.
Ursolic acid		-8.54	H- bond: ARG 493. H-I: ARG 403, TYR 453, SER 494, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505.
Withaferin A		-7.53	H- bond: TYR 449 and SER 496. H-I: ARG 403, TYR 453, ARG 493, SER 494, TYR 495, PHE 497, TYR 501 and HIS 505.
Withanone		-8.12	H- bond: ARG 403, SER 494 and SER 496. H-I: TYR 449, TYR 453, ARG 493, TYR 495, PHE 497 and HIS 505.

Withanolide		-7.73	H- bond: SER 494 and SER 496. H-I: ARG 403, TYR 453, ARG 493, TYR 495, PHE 497, TYR 501 and HIS 505.
Zeylanone		-7.21	H- bond: ARG 493 and SER 496. H-I: TYR 453, SER 494, TYR 495, PHE 497, TYR 501 and HIS 505.

From Table 2, we can observe that the binding energy of all the 15 selected phytochemicals was higher (≤ -7.00 kcal/mol) than the HCQ which was chosen as the std. reference drug to study and compare the binding affinity of these phytochemicals against the spike protein.

The binding energy of standard drug HCQ was found to be -5.22 kcal/mol whereas, Glycyrrhetic acid showed the highest binding energy among all 15 phytochemicals with the value of -9.02 kcal/mol towards the active site of the target protein, followed by Oleanolic acid with the value of -8.68 kcal/mol, Ursolic acid -8.54, Maslinic acid -8.17, Withanone -8.12, Withanolide -7.73, Oleanane -7.63, Withaferin A -7.53, Betulinic acid -7.46, β- sitosterol -7.45, Kamolonol -7.41, Foetidin -7.36, Zeylanone -7.21, Marrubiin -7.09 and Desmodin which showed the lowest binding energy of value -7.07 kcal/mol. The binding of the receptor protein (7T9J_C) with each ligand (phytochemical) is represented in Figure 5(a)-20(a). Protein-ligand interactions of each phytochemical were estimated by Ligplot+ 2.2 software and are represented in Figure 5(b)-20(b).

ARG 403, ARG 493, ARG 498 and HIS 505 were the four amino acid residues involved in the hydrogen bond with HCQ. TYR 449, TYR 453, SER 494, TYR 495, SER 496, PHE 497 and TYR 501 were the amino acids involved in hydrophobic interactions with the spike protein. Glycyrrhetic acid formed hydrogen bonds with the TYR 453 and SER 494. ARG 403, ARG 493, TYR 495, SER 496, TYR 501 and HIS 505 were the amino acid residues of spike protein that interacted with Glycyrrhetic acid by hydrophobic interaction. Similarly, Oleanolic acid showed the hydrogen interaction with the ARG 493 whereas, ARG 403, TYR 453, SER 494, TYR 495, SER 496, TYR 501 and HIS 505 showed the hydrophobic interaction with the amino acid residues of the spike protein. ARG 493 amino acid residue of spike protein was involved in hydrogen bonding with the Ursolic acid. Besides, ARG 403, TYR 453, SER 494, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505 were involved in hydrophobic interaction with the targeted site of the protein. Maslinic acid formed a hydrogen bond with the ARG 493. In addition to this, ARG 403, TYR 453, SER 494, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505 were the amino acids involved in hydrophobic interactions with the spike protein. Withanone showed hydrogen bonding with ARG 403, SER 494 and SER 496 amino acid residues and moreover interacted with TYR 449, TYR 453, ARG 493, TYR 495, PHE 497 and HIS 505 amino acids residues by hydrophobic interactions. Withanolide formed hydrogen bonds with the SER 494 and SER 496 residues. ARG 403, TYR 453, ARG 493, TYR 495, PHE 497, TYR 501 and HIS 505 amino acid residues of spike protein interacted with Withanolide by hydrophobic interaction. Oleanane showed the hydrophobic interaction with the ARG 403, ARG 493, SER 494, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505 amino acid residues of the spike protein, whereas it didn't show hydrogen interaction with the any of the amino acid residues of the spike protein. Withaferin A showed hydrogen bonding with TYR 449 and SER 496. Moreover, it interacted with ARG 403, TYR 453, ARG 493, SER 494, TYR 495, PHE 497, TYR 501 and HIS 505 amino acid residues via hydrophobic interaction. The Betulinic acid formed a hydrogen bond with the ARG 493. ARG 403, TYR 453, SER 494, TYR 495, SER 496, PHE 497 and HIS 505 showed the hydrophobic interaction with the amino acid residues of the spike protein.

Similarly, β-sitosterol showed the H- bond with HIS 505, whereas, hydrophobic interactions with the ARG 403, TYR 453, ARG 493, SER 494, TYR 495, SER 496, PHE 497 and TYR 501 amino acid residues with the targeted protein site. TYR 453 and SER 494 residues of spike protein were involved in H-bond formation with Kamolonol. Besides, ARG 403, ARG 493, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505 residues showed the hydrophobic interactions with the spike protein. Foetidin formed H-bond with ARG 493 amino acid residue. Moreover, Foetidin interacted with the ARG 403, TYR 449, TYR 453, SER 494, TYR 495, SER 496, PHE 497, TYR 501 and HIS 505 residues hydrophobically. Zeylanone showed hydrogen bonding with ARG 493 and SER 496. In addition to this, Zeylanone interacted with TYR 453, SER 494, TYR 495, PHE 497, TYR 501 and HIS 505 amino acid residues via hydrophobic interaction. Similarly, Marrubiin formed a hydrogen bond with the SER 496 amino acid residue. Furthermore, it interacted with the ARG 403, TYR 453, SER 494, TYR 495, PHE 497, TYR 501 and HIS 505 residues hydrophobically. The Desmodin formed a hydrogen bond with the SER 494 amino acid residue. In addition to this, Desmodin interacted with the ARG 403, TYR 449, TYR 453, ARG 493, TYR 495, SER 496, PHE 497 and HIS 505 residues hydrophobically.

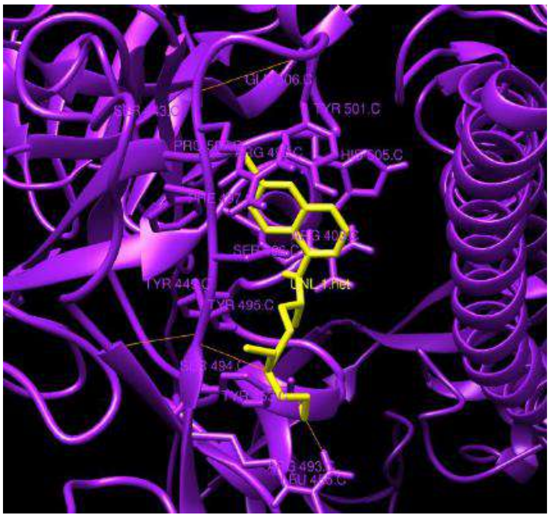


Fig.5(a): 7T9J_C - HCQ

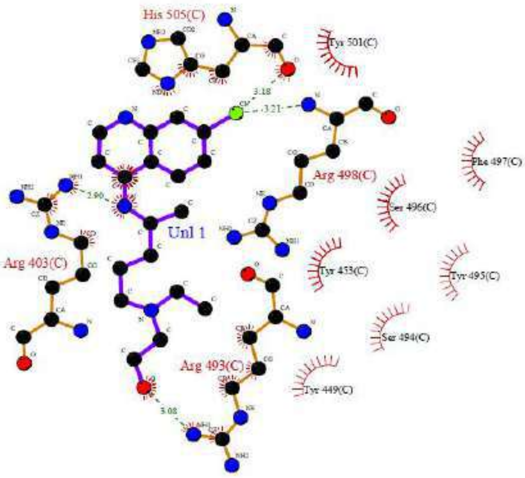


Fig.5(b): 7T9J_C - HCQ

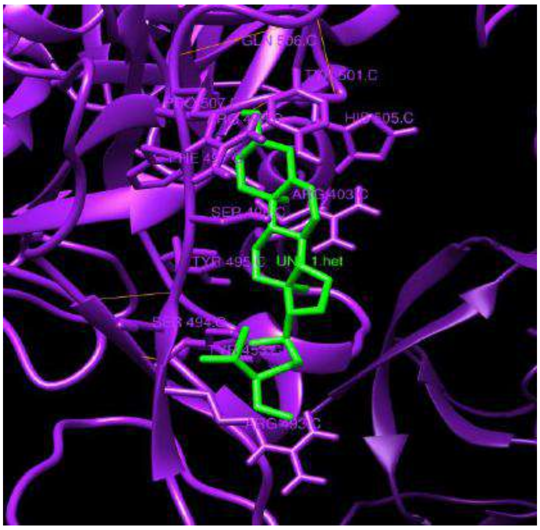


Fig. 6(a): 7T9J_C - β- Sitosterol

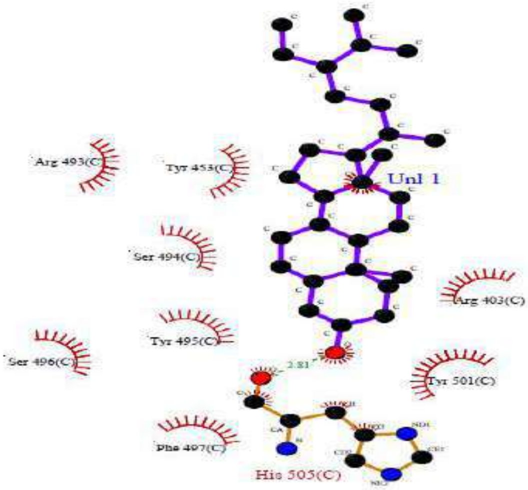


Fig. 6(b): 7T9J_C - β- Sitosterol

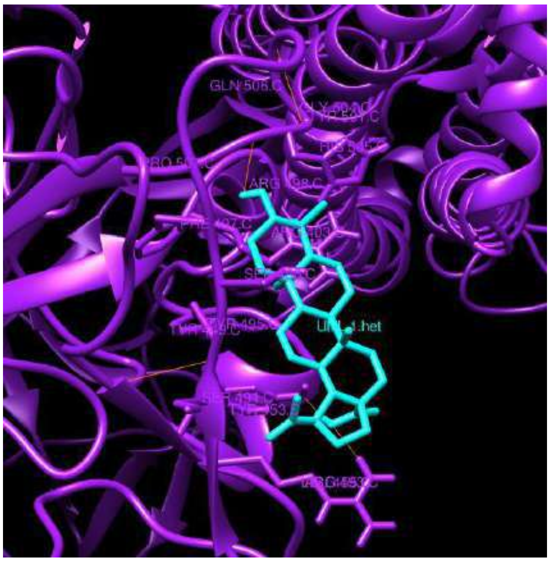


Fig. 7(a): 7T9J_C - Betulinic acid

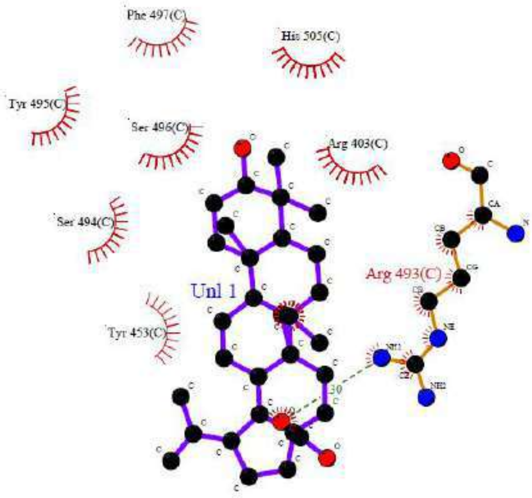


Fig. 7(b): 7T9J_C - Betulinic acid

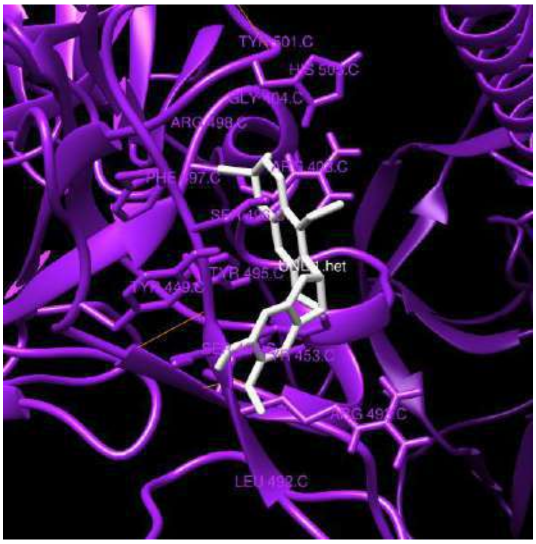


Fig. 8(a): 7T9J_C - Desmodin

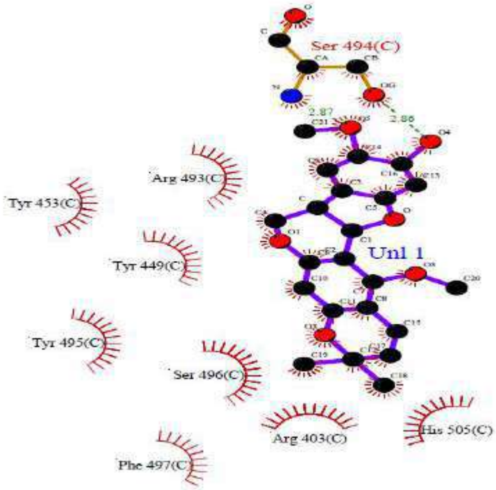


Fig. 8(b): 7T9J_C - Desmodin

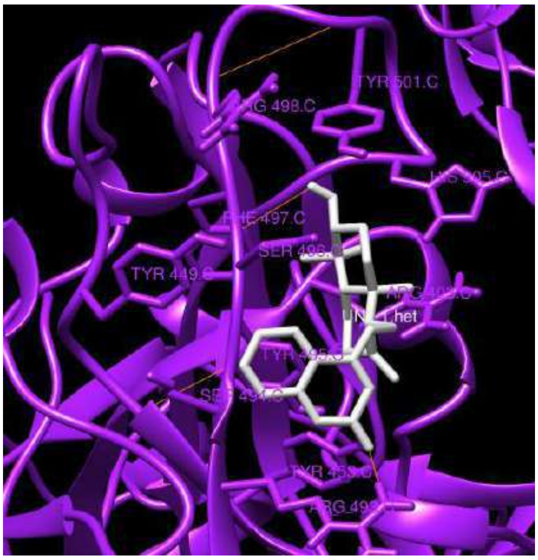


Fig. 9(a): 7T9J_C – Foetidin

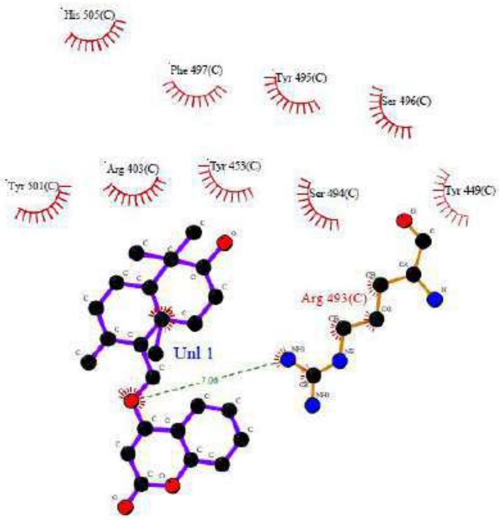


Fig. 9(b): 7T9J_C - Foetidin

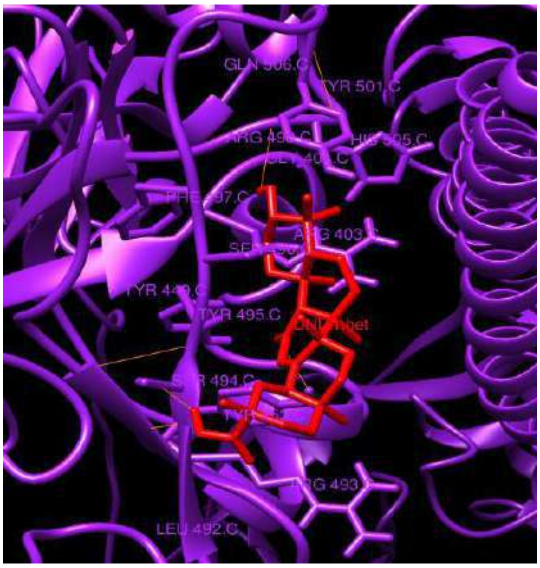


Fig. 10(a): 7T9J_C - Glycyrrhetic acid

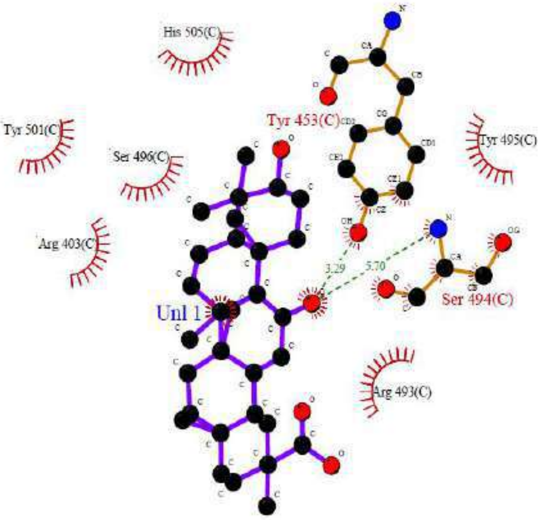


Fig. 10(b): 7T9J_C - Glycyrrhetic acid

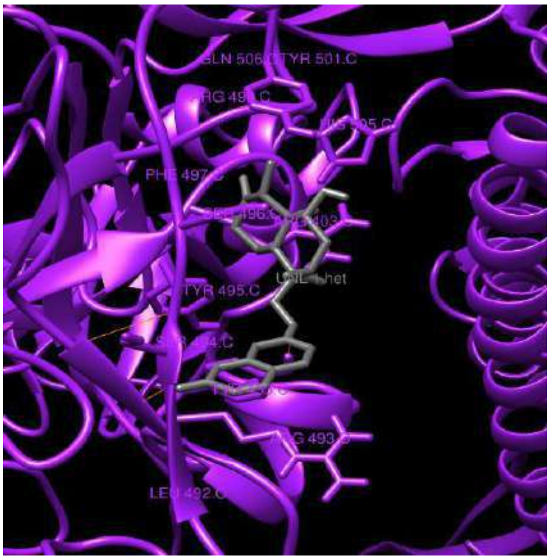


Fig. 11(a): 7T9J_C - Kamolonol

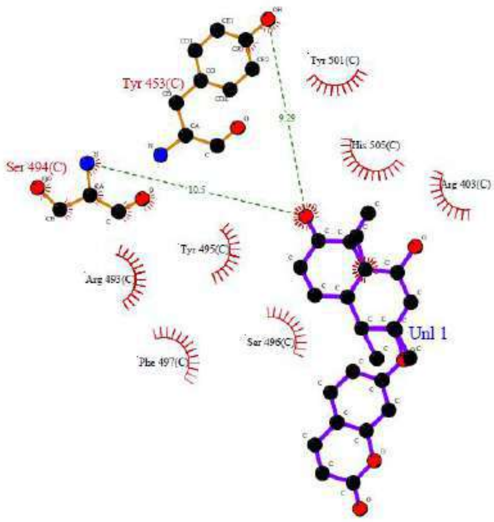


Fig. 11(b): 7T9J_C - Kamolonol

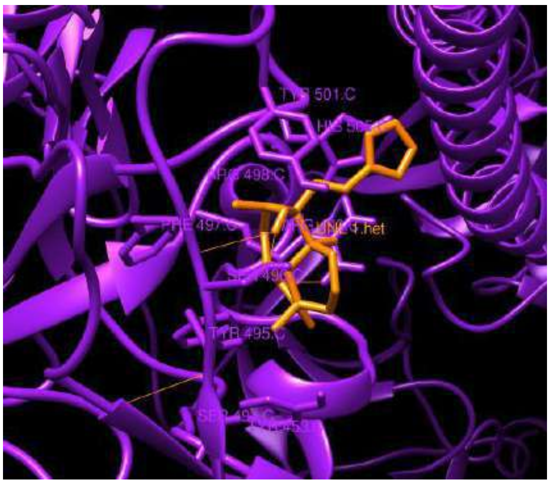


Fig. 12(a): 7T9J_C - Marrubiin

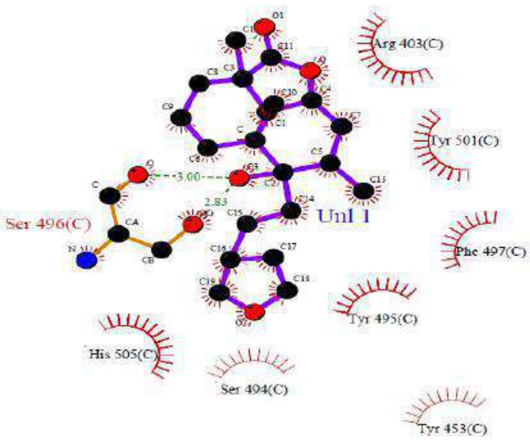


Fig. 12(b): 7T9J_C - Marrubiin

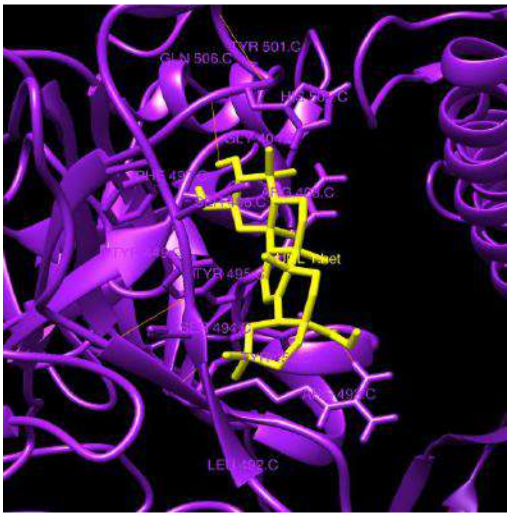


Fig. 13(a): 7T9J_C - Maslinic acid

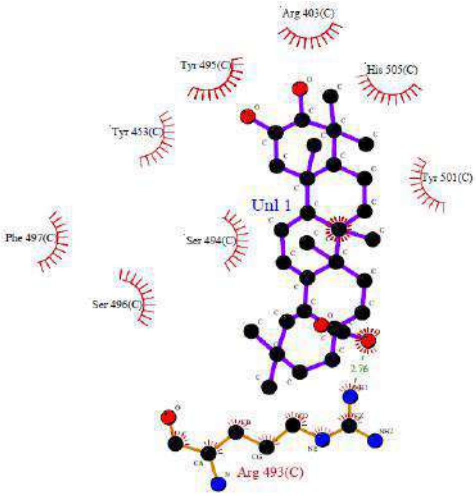


Fig. 13(b): 7T9J_C - Maslinic acid

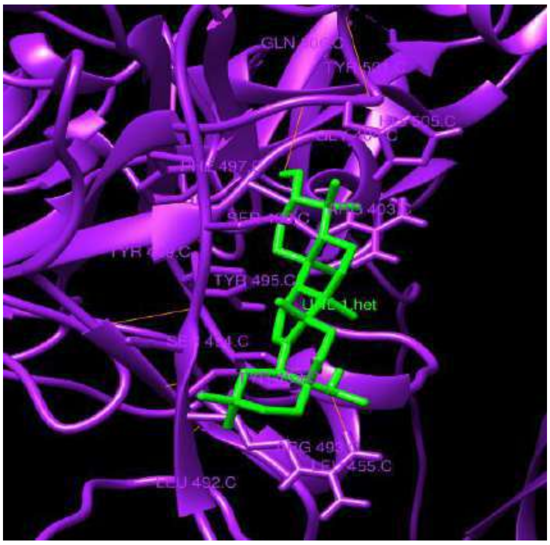


Fig. 14(a): 7T9J_C - Oleanolic acid

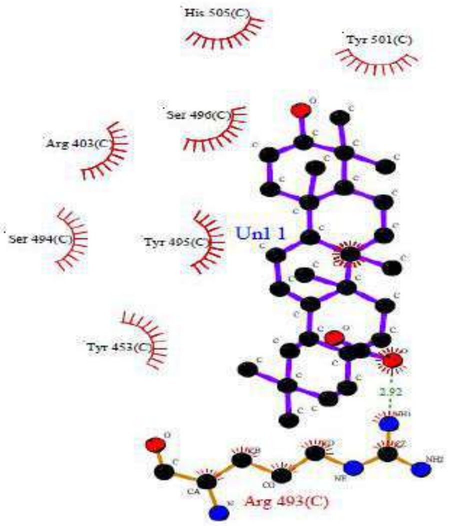


Fig. 14(b): 7T9J_C - Oleanolic acid

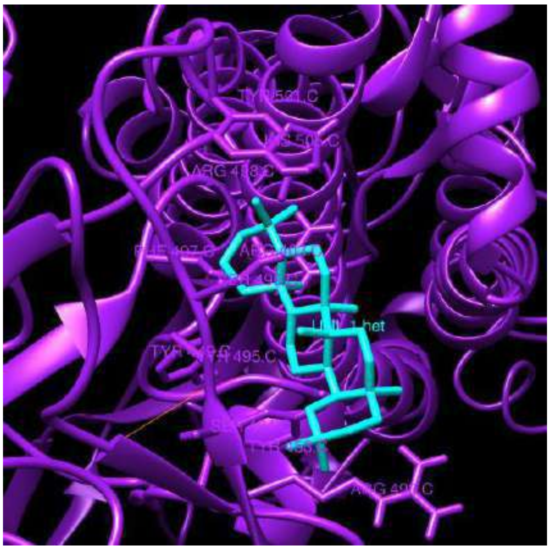


Fig. 15(a): 7T9J_C – Oleanane

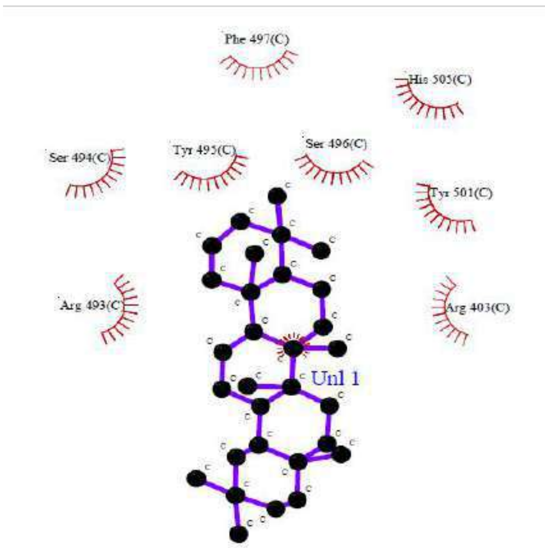


Fig. 15(b): 7T9J_C – Oleanane

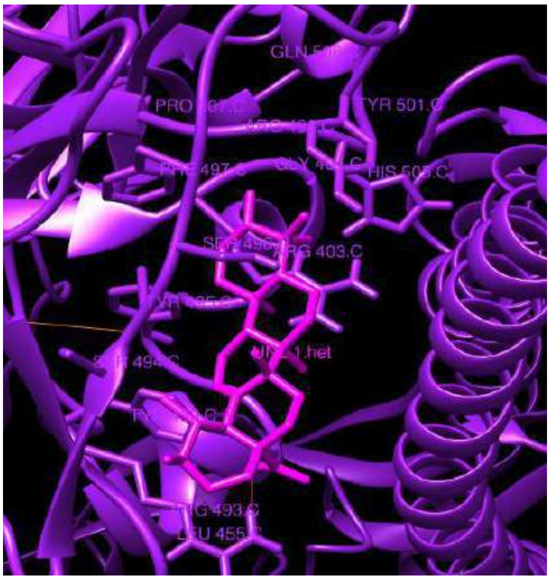


Fig. 16(a): 7T9J_C – Ursolic acid

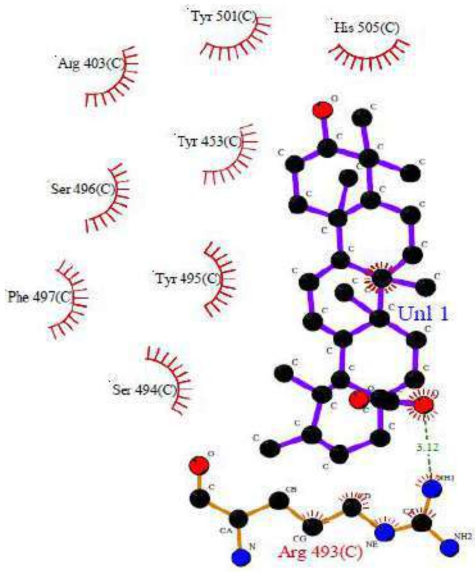


Fig. 16(b): 7T9J_C – Ursolic acid

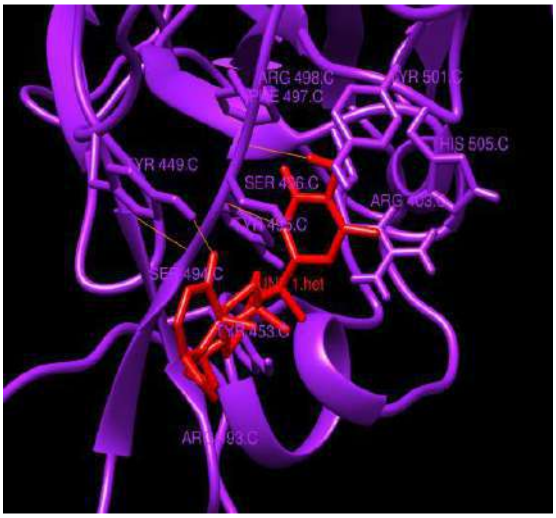


Fig. 17(a): 7T9J_C – Withaferin A

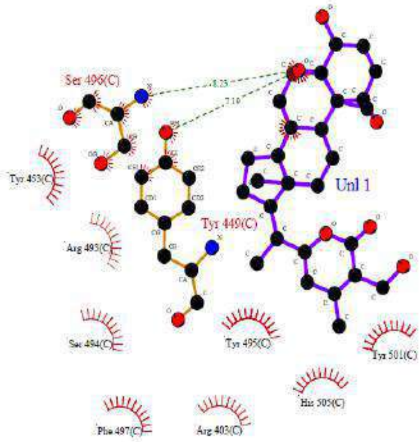


Fig. 17(b): 7T9J_C – Withaferin A

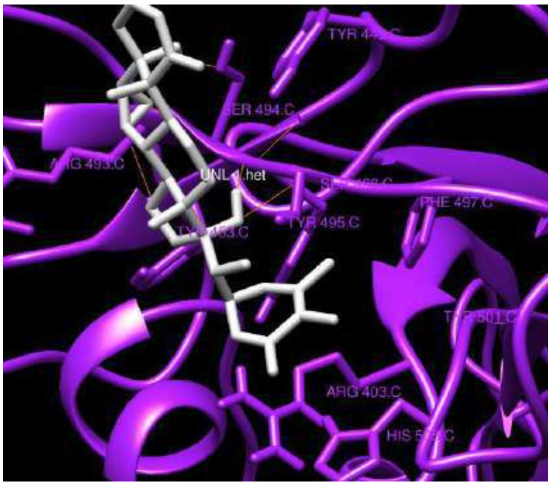


Fig. 18(a): 7T9J_C – Withanone

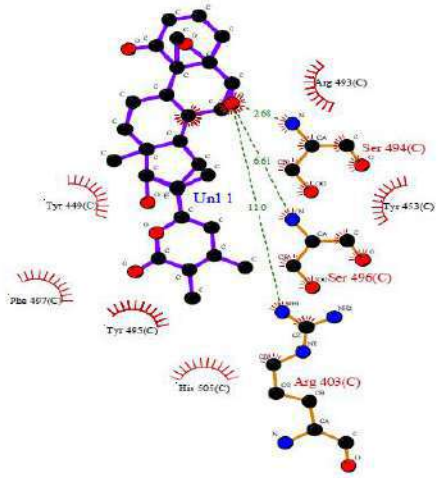


Fig. 18(b): 7T9J_C – Withanone

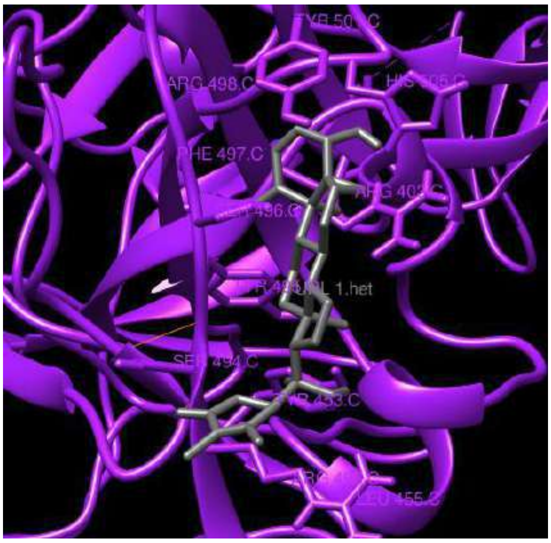


Fig. 19(a): 7T9J_C – Withanolide

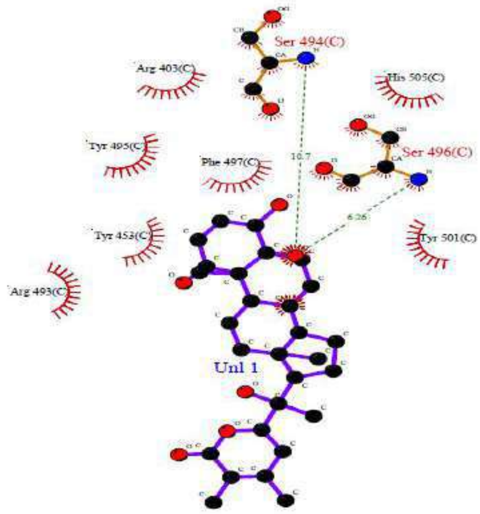


Fig. 19(b): 7T9J_C – Withanolide

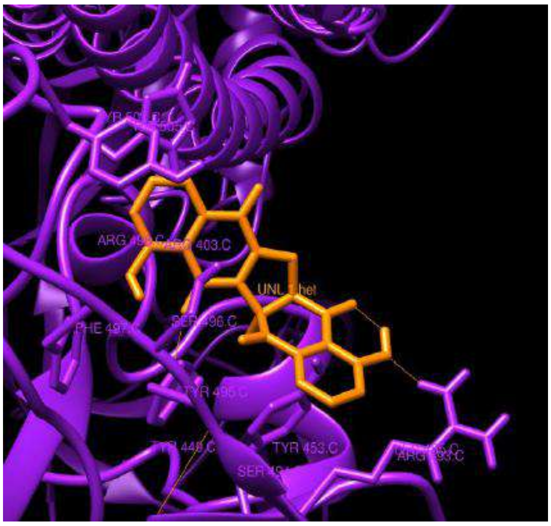


Fig. 20(a): 7T9J_C – Zeylanone

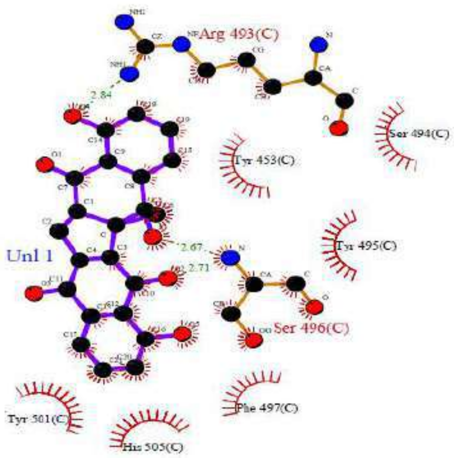


Fig. 20(b): 7T9J_C – Zeylanone

Fig.5(a)-20(a): Crystal structure of the protein receptor-binding domains (7T9J_C) and ligand complexes of HCQ, β-Sitosterol, Betulinic acid, Desmodin, Foetidin, Glycyrrhetic acid, Kamolonol, Marrubiin, Maslinic acid, Oleanolic acid, Oleanane, Ursolic acid, Withaferin A, Withanone, Withanolide, and Zeylanone.

Fig.5(b)-20(b): Hydrogen and hydrophobic interactions of all the compounds with the spike protein of the omicron variant (7T9J_C).

3.2 ADME studies & Drug likeliness prediction

Besides, the calculating the binding energy and the molecular interactions between the protein and the ligand, compounds were further allowed for ADME analysis. These properties evaluate the ability of a compound to act as a drug, which follows Lipinski's Rule of Five.

Lipinski's rule of five is a rule of thumb that describes the drug likeliness of a compound, with specific therapeutic properties that are likely to have chemical and physical properties that would make it an orally active drug. This rule figures out a few of the molecular properties that are essential for the drug pharmacokinetics in the human body, such as Absorption, distribution, metabolism, and excretion.

Lipinski's rule of five states that the chemical compound must follow the below-mentioned criteria to be considered a safe drug,

1. Molecular mass > 500 Da
2. A logarithm octanol-water partition coefficient (MlogP) > 4.15

3. Number of H-bond donors > 5
4. Number of H-bond acceptors >10

The violation of 2 or more conditions anticipates that the compound is not orally active [71]. Swiss ADME web server was used to study the ADME properties of the phytochemicals. Table 3 shows the ADME analysis data obtained from the SwissADME server for all the 15 phytochemicals along with HCQ.

Table 3: ADME properties of the few selected compounds

Name of the Phytochemical	Molecular Weight (<500 Da)	MlogP (<4.15)	H – Bond Acceptor (<10)	H – Bond Donor (<5)	Violations	Drug Likeliness
Hydroxychloroquine	335.87	2.35	3	2	0	Yes
β- Sitosterol	414.71	6.73	1	1	1	Yes
Betulinic acid	456.70	5.82	3	2	1	Yes
Desmodin	382.41	2.06	6	1	0	Yes
Foetidin	382.49	3.83	4	1	0	Yes
Glycyrrhetic acid	470.68	4.87	4	2	1	Yes
Kamolonol	398.49	3.00	5	1	0	Yes
Marrubiin	332.43	2.76	4	1	0	Yes
Maslinic acid	472.70	4.97	4	3	1	Yes
Oleanolic acid	456.7	5.82	3	2	1	Yes
Oleanane	412.73	9.05	0	0	1	Yes
Ursolic acid	456.70	5.82	3	2	1	Yes
Withaferin A	470.6	2.75	6	2	0	Yes
Withanone	470.6	2.75	6	2	0	Yes
Withanolide	470.6	2.75	6	2	0	Yes
Zeylanone	374.34	0.69	6	2	0	Yes

From Table 3, we can observe that all the 15 phytochemicals followed the Lipinski rule of five. Out of which, 8 phytochemicals including Desmodin, Foetidin, Kamolonol, Marrubiin, Withaferin A, Withanone, Withanolide, and Zeylanone showed no violations. And the remaining 7 phytochemicals including β- Sitosterol, Betulinic acid, Glycyrrhetic acid, Maslinic acid, Oleanolic acid, Oleanane, and Ursolic acid showed one violation (MlogP < 4.15) in Lipinski rule of five.

The data obtained from Table 3, shows that all the selected phytochemicals followed the Lipinski rule of five with not more than 1 violation, making them a suitable drug candidate for the treatment against the omicron variant.

The current study was focused on identifying the inhibitory potential of the 15 selected phytochemicals by targeting the active sites of SARS CoV-2 omicron variant spike protein. This protein was chosen as the druggable target site, since it plays a crucial role in the recognition, attachment, and fusion of the virus into the host cell. There are several chemotherapeutic drugs such as Remdesivir, Azithromycin, Amantadine, and many more that are being used to treat the infection. But, few of the drugs showed certain side effects like low blood pressure, vomiting, nausea, orthostatic hypotension, allergic reactions, and others. In addition to this, higher uptake of chemotherapeutic drugs might cause severe toxic effects on the human body that can lead to organ dysfunction.

Hence, there is an urgent need for developing an effective drug or inhibitor against the omicron variant with the least toxic effects. As a consequence of that, these phytochemicals can be the best solution for the treatment, whose binding energies were found to be 1.5-1.8 times higher than the std. HCQ, following the Lipinski rule of five criteria and are known to be safer with no or fewer side effects, thus intimating that these phytochemicals have the potential to form an antiviral drug or inhibitor against the SARS CoV-2 omicron variant.

IV. CONCLUSION

The mutating nature of the SARS CoV-2 has resulted in the emergence of omicron; another new variant with a higher fatality rate has created urgency for the development of new antiviral drugs or vaccines against the infection. In this regard, the research study was carried out, to identify the therapeutic potential of the phytochemicals from different medicinal plants against the spike protein of the omicron variant using Insilico approaches. This study showed that the binding energy of all the 15 phytochemicals was higher compared to the std. HCQ with the greater potential of inhibiting the binding of the viral spike protein to the host cell receptor. In addition to this, the phytochemicals also fulfilled all the criteria of the drug likeliness and ADME studies. Therefore, the results obtained from the current study suggest that all these 15 phytochemicals can be used as lead molecules for designing an effective drug against the omicron variant.

However, the data obtained from the current study is based on the Insilico approach, additional studies have to be carried out with in vitro and in vivo conditions using animal models to check the feasibility of the compound.

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Proximate Analysis of Apu-Apu Leaf Weeds in Lake Tondano Waters as Raw Material for Tilapia (Orechromis niloticus) Feed Formulation

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Abstract—*The purpose of this study was to analyze the nutritional content (Proximate Test) of Apu-apu Leaf weeds in the waters of Lake Tondano and to determine the growth of Tilapia (Oreochromis niloticus) fry that consume pellets of raw material for the formulation/composition of Apu-apu Leaf (Pistia stratiotes) in different percentage levels. This research is one solution to obtain raw materials for fish feed from weeds in Lake Tondano so that it can overcome the high cost of feed and minimize pollution in Lake Tondano. The general approach used to achieve the objectives of this research activity is the collection from Lake Tondano, namely the leaves of Apu-apu (Pitsia stratiotes). The test fish were obtained from fish cultivators in Eris Village, Eris District, Minahasa Regency. Fish size 5-8 cm. Prior to the research, the fish were acclimatized for one week for environmental adjustment. The study was carried out by following a 5 treatment design and 3 replications, the design used was RAL, and measurements of ratios, daily growth and feed efficiency values. The analytical method applied is proximate analysis, carried out at the Baristand Manado laboratory. This research produces outputs in the form of scientific works, namely International Journals. The results showed that treatment C gave the highest relative growth (168.99%) followed by treatment A (93.28 %), D (84.73), E (83.81%) and B (44.65%). The highest feed efficiency value was also shown by treatment C (38.01%) then treatment A (24.68%), D (22.59%), E (22.19%) and B (13.87%).*

Keywords— ***Proximate Analysis, Apu-apu Leaf, Fish Feed, Tondano Lake.***

I. INTRODUCTION

Lake Tondano is an asset owned by the Province of North Sulawesi, which is one of the places for community livelihoods. The current existence of Lake Tondano is very concerning because of the occurrence of eutrophication. Eutorfication is the enrichment of nutrients that results in the occurrence of aquatic weed blooms. The definition of weed is a plant that grows around waters whose presence is not desired in the vicinity of the cultivation activities carried out. The livelihoods of the people in Lake Tondano are fish farming activities with a floating net cage system, where fish cultivation in Lake Tondano is currently no longer going well, this situation is caused by contamination of water weeds on the surface and in water bodies. The growth of aquatic weeds in Lake Tondano causes the placement of space for fish cultivation to be narrow and overall greatly affects the quality of water for ideal fish life needs (Korah, 2000).

The phenomenon of Lake Tondano no longer looks ideal in terms of multi-functions (hydropower, PDAM, fisheries, agriculture, animal husbandry and aesthetics). This condition describes a situation that must be found a solution to minimize pollution in Lake Tondano, so that the multi-function of Lake Tondano can be carried out according to its designation. Weeds (Apuapu leaves) that grow rapidly in water bodies and on the water surface of Lake Tondano can be reduced by utilizing these weeds into a useful product, especially for the fisheries sector, which can be used as raw material for fish feed (Tamanampo et al., 1995)

Increasing fish production is highly dependent on the consumption of feed for cultured fish, but fish feed is an obstacle for cultivators. The obstacle faced by cultivators is the increasing price of feed. Fish farming activities will incur costs for feed 70% of a maintenance cycle, therefore the use and feeding must be carried out effectively and efficiently (Kusen, 2014).

II. MATERIAL AND METHODS

The target of this research is the collection of Apu-Apu Leaf weeds where the object of sampling is Lake Tondano. The number of Apu-apu leaves to be taken is according to research needs.

1. Stages of Collection and Treatment of Apu-Apu-Leaf Weeds

Weeds are taken on the surface of the lake water, in areas that are overgrown with apu-apu leaves and selected that physically look fresh. Weeds were taken as needed in fish feed formulations for fish rearing during the research period. Treatment of weeds are: (1). Washed clean to remove dirt attached to the leaves and roots removed; (2). Cut into small pieces; (3). Dried in the sun and if it is still not dry can be continued drying in the oven; (4). After drying, the weeds were blended into fine powder or powder and prepared as much as 200 grams for analysis of the nutritional content (Proximate test) in the laboratory. Treatment for fish flour, soy flour, coconut flour is made by grinding the ingredients, soybeans and coconut so that they become flour-like shapes.

2. Stages of Preparation for Trial of Feed Composition on the Growth of Tilapia (Oreochromis niloticus) Using Apu-Apu Leaves (Pistia stratiotes) as raw materials

The maintenance container is using a plastic pan with a diameter of 56 cm and a height of 30 cm as many as 15 pieces, each filled with 10 liters of fresh water. The tools used are: aerator for oxygen source, thermometer for measuring temperature, litmus paper for measuring pH, digital scales, besides that, a set of tools for making feed is also provided (grinder, sieve, spoon, tray, plastic pan and electric oven).

3. Stages of Experiment Implementation

Each container is stocked with 10 tilapia fish and the initial weight is weighed, The frequency of feeding 3x a day at 07.00, 12.00 and 17.00 with a dose of 5% of body weight, Adjustments to the feeding of the test fish were carried out every 2 weeks and the test fish were weighed using a digital scale. Temperature measurements are carried out once a week while pH measurements are carried out before and after water changes. Siphoning of feces in containers is carried out every day by using a plastic hose to remove leftover feed and feces at the bottom of the container. Water changes were carried out 2 times a day by replacing of the water from the research container. The measurement of fish weight was carried out at an interval of 2 weeks and the measurement time was for 2 months.

4. Research design

Treatment Design: there were 5 treatments that were tested and each treatment was replicated 3 times thus there were 15 experimental units, the treatments being tested were:

1) Treatment A : Pellets without Buffalo Leaves as a Control,
2) Treatment B: Pellets with 10% Apu-apu Leaf flour,
3) Treatment C: Pellets with 20% Apu-apu Leaf flour,
4) Treatment D: Pellets with 30% Apu-apu Leaf flour,
5) Treatment E: Pellets with 40% Apu-apu Leaf flour.

Environmental Design: The design used is a Completely Randomized Design (CRD) which is based on the assumption that all experimental units and environmental conditions are homogeneous with a mathematical model:

Response Design: Parameters measured were relative growth, daily growth and feed efficiency value. The growth observed was the growth of fish body weight where the measurement was using a scale. The analyzed variables are: Relative Growth (%) how to calculate it by using the formula (Weatherley, 1988).

III. RESULTS AND DISCUSSION

The results of observations on the growth of tilapia seeds during the study period showed a good response where the feed given could be consumed. The presence of a reduction in the amount of Apu-Apu Leaf flour contained in the test feed (A, B, C, D and E) had an effect on the amount of growth that could be achieved by each test fish, so that at the end of the study, differences in relative growth and feed efficiency value. The results of the calculation of the weight growth obtained from each time the fish fry are weighed the value required in Appendix 2. The calculation of the relative growth achieved by each treatment is shown in Appendix 3. The amount of feed given during the study is shown in Appendix 4. Summary of the effect of the use Apu-Apu leaf meal flour on the growth of tilapia fry and the efficiency value of the test feed is shown in Table 1.

Table 1. Growth of Fish Seed Values Given Apu-Apu Leaf Starch Feed and Efficiency Value of Each Test Feed

Treatment	Average Weight (gr)		Growth (%)		Feed Consumption	
	First	And	Nisbi	Daily	Total (gr)	Feed Efficiency (%)
A	34.7	67.0	93.28	4.71	130.90	24.68
B	34.3	49.7	44.65	2.64	111.07	13.87
C	34.3	92.3	168.99	7.07	152.60	38.01
D	34.7	64.0	84.73	4.36	129.73	22.59
E	35.0	64.3	83.81	4.35	132.07	22.19

The highest daily growth rate was shown by fish that received treatment C and the lowest was found in fish that received treatment B. The temperature and pH of the water, which are water quality parameters measured in each container during the study, were in a suitable range for the life of tilapia fry. water temperature was recorded between 26-27ºC and water pH between 6.5-7.5.

1. Relative Growth

Tilapia seeds that were tested on different feeds in the amount of Apu-Apu Leaf flour contained therein showed a response to the growth of each treatment with changes in time. The results of the calculation of the relative growth which is the response of each treatment are then analyzed by calculating the analysis of variance from the results of these calculations, which are then described in Table 2.

Table 2. Analysis of Various Effects of Treatment on Relative Growth

Diversity	DB	JK	KT	Fcount	F_{table}	
					0.05	0.01
Treatment	4	4391.01	1097.753	16.915	3.478	5.994
Error	10	648.96	64.896			
Total	14	5039.98				

Based on the results of the analysis of variance in Table 2 above, it can be seen that the value of Fcount (16.915) is greater than the value of Ftable (3.478-5.994). This gives an answer that the experimental treatment has a significant effect on the relative growth of tilapia fry. This also shows that there is a different effect between the treatments being tested. To find out more about the difference between each treatment, Duncan's multiple-area test was conducted.

Based on the results of Duncan's multiple-area test, where the comparison of the average values showed that treatment A (pellets without apu-apu leaf powder) and treatment C (pellets with 20% hibiscus leaf powder had the same effect (not significant) on relative growth. , the percentage of growth obtained was higher than other treatments (B, D, and E). This illustrates that the use of apu-apu leaf meal (20%) will provide a good relative growth compared to the use of apu-apu leaf meal which is slightly or too much. The use of 10% and without Apu-Apu Leaf flour showed results in the form of an insignificant effect on the relative growth gain, this means that the amount of fish meal replaced by Apu-Apu Leaf flour, although reducing the amount of protein content, has not shown any significant effect. The effect is because the range of protein requirements by tilapia fry is still below the range in feed B (27.59%). Treatments E and B gave the same effect on the acquisition of the relative growth rate, this means that the amount of Apu-apu leaf meal contained in the feed formulation for tilapia was 40 and 10%, the effect was still higher (83.81%) compared to the treatment B (44.65). The low achievement of the two treatments was possible because the percentage of fish meal replaced by the use of apu-apu leaf meal (40 and 20%) had an impact on reducing the percentage of protein content in the test feed (13.51-27.59).

The best results from treatment C with a protein content of 23.07% were also shown by Lapadi (2001) where the use of feed containing 30.19% protein which was mixed from caterpillar flour from oil palm waste gave a relative growth of 598.68% for tilapia size 13-15 grams / head. Furthermore, Luquet (1991) added that value fish will grow well if the food contains 29% protein. To find out more about the relative growth response that has been achieved during the study, it can be seen in Figure 4. In the observation of the weight gain of tilapia fish which were weighed every two weeks, it was seen that there was a difference in growth rate where treatment C at each weighing showed the highest achievement rate (from the first development to fourth), this is because all the feed given can be used for growth. In the second weighing the relative growth shown by treatments D and E was the same (49.13%), this was because the amount of energy in the two test feeds was not so different (430.45) and 420.49 calories/gr). Treatments D and E were always on the lowest graph because they were influenced by the insufficient nutrient content in the feed to stimulate growth. There were two main factors that caused the low growth rate of treatment E, the first was the very low protein content, which was only 13.51% and the second was the lack of energy (420.49%). In the third measurement (sixth week) it was seen that there was a difference between treatments A and D, where treatment A showed a higher growth rate than treatment D. This was because the amount of feed consumed by the test fish that received treatment A was more than with treatment D where in treatment D it was found that there were food remnants that were not eaten by the test fish. Likewise, the fourth measurement of treatment A was greater than treatment B where the feed given to treatment B could not be consumed entirely by the test fish, this was because during the sixth week the appetite of the test fish decreased.

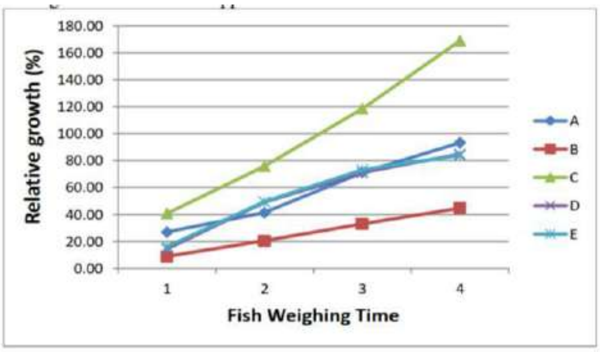

Fig.1. Test Fish Relative Growth Chart

From Figure 1 it is clear that the increase in fish weight with changes in maintenance time. The mapping of the data on the highest relative growth of tilapia seeds was given by treatment C (168.99%) for eight weeks of rearing when compared to other treatments. The results of the proximate analysis showed that the protein content decreased along with the increase in the ApuApu Leaf flour that made up the pellet formulation. From the proximate results of the test feed in Table 2, it shows that treatment A without butterflyfish leaves contains 38.65% protein, treatment B with pellets composed of 10% butterflyfish leaf, 27.59% protein, treatment C with 20% composition, 23 protein. 0.07%, treatment D with a composition of 30%; protein 22.76% and treatment E with a composition of 40%; 13.51% protein. When viewed from treatment A, the protein content was higher than the protein possessed by treatments B, C, D, and E. It appears that the higher the addition of Apu-Apu Leaf flour will decrease the protein content, this is because Apu-Apu Leaf flour is not a good source of protein. major in feed.

The large variety of raw materials used in the preparation of the formulation of a pelletshaped feed determines the quality of the feed, because the more varied the use of raw materials, it is clear that the amino acid content that composes the protein is more complete, as well as other elements such as fats, carbohydrates, vitamins and minerals. In this study, tilapia is a herbivorous type of fish, so it can be seen that although the use of fish meal in treatment A was much higher than that in treatment C, it seems that tilapia has good growth with the addition of 20% Apu-Apu Leaves, this can be seen from received response to the growth of tilapia. Furthermore, Djajasewaka (1985), stated that to get good growth is not only determined by high protein but also by supporting substances such as fat, carbohydrates, vitamins and minerals.

2. Feed Efficiency Value

Data on the value of feed efficiency from each treatment that was tested during the 8-week rearing period can be seen in Appendix 3 and the results of the analysis of variance on feed efficiency can be seen in Table 3.

Table 3. Analysis of Various Effects of Treatment on the Value of Feed Efficiency

Diversity	DB	JK	KT	F_{count}	F_{tabel}	
					0.05	0.01
Treatment	4	104.55	26.137	57.910**	3.478	5.994
Error	10	4.51	0.451			
Total	14	109.06				

Based on the results of the analysis of variance in Table 3, it can be seen that Fcount is greater than Ftable at the 1% level. This means that there is a difference between the treatments being tested. On this basis, further tests were carried out with Duncan's multiple region test. The test results can be seen in Appendix 8, where treatment C was significantly different from treatment B, D, and E, and not significantly different from treatment A. Furthermore, treatment A was significantly different from treatment D and E, and significantly different from treatment B. While treatments B, D, and E each gave the same effect (not significant). To determine the response trend indicated by the value of feed efficiency mapped through the histogram in Figure 5.

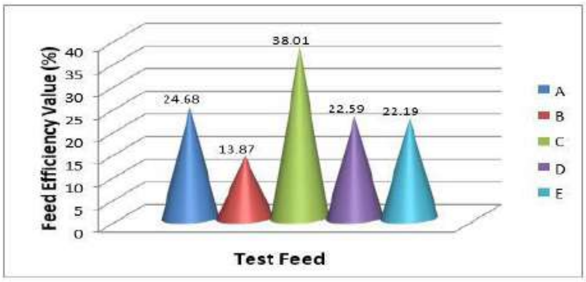

Fig.2. Histogram of Test Feed Efficiency Value (%)

Figure 2 above, shows the feed efficiency of each treatment. Treatment C gave the highest feed efficiency value (38.01%) when compared to other treatments, while treatment B was the lowest (13.87%) because treatment C with 20% Apu-apu leaf meal gave a taste or aroma favored by fish. test so that the fish's appetite increases, besides that tilapia only requires 25-30% protein so that the efficiency value of the test feed gives a good response. Feed has a function as energy that supports fish growth, therefore it is necessary to provide quality and efficient feed. Djajasewaka (1985), stated that a good feed is expected to contain 20-60% protein, 4-18% fat and 10-50% carbohydrates. Carbohydrates consisting of BETN and crude fiber greatly affect the efficiency or not of the feed given to fish. BETN is the part of carbohydrates that can be digested by fish while crude fiber is the part that is difficult for fish to digest in producing energy. Thus, the greater the crude fiber in the feed, the less favorable the fish growth, the higher the BETN in the feed, the more efficient the feed will be.

The efficient value of feed aims to determine whether the feed being tested is efficient or not, in the sense of whether the feed provided is used properly by fish and provides growth or not. According to Djarijah (1995), feed efficiency indicates the quality of the feed. Where the greater the value of the efficiency of eating, the higher the quality of the feed. Conversely, the smaller the efficiency value means the lower the quality of the feed. The use of Apu-Apu Leaf flour as much as 20% has given the highest efficiency value among treatments, the efficiency value is quite high when compared to the results achieved by the use of kale leaf meal which was tested on tilapia fish by Marthen (1999) with an efficiency value highest (49.43%), on the other hand Timburas (2000) found that the feed efficiency value of water hyacinth leaf meal was the best among the feeds tested on tilapia, which was 40.31%. while the use of water hyacinth leaf flour given to red tilapia gave a feed efficiency value of 53.05% (Girsang, 1999), using tapung cab eke in papaya grain flour pellets got an efficiency value of 176.06 (Rau, 1990). The use of corn cobs flour as a substitute for fish meal in the diet formulation for the growth of red tilapia gave the best efficiency value of 43.28% (Onibala, 1995). Meanwhile, the use of seagrass meal on the growth of red tilapia gave an efficiency value of 23.24% (Kumean, 1998). Furthermore, Mandagi (2003) obtained an efficiency value of 66.59% using noni leaf meal as fish seed feed and the use of green algae flour in tilapia seed feed gave feed efficiency of (41.81% (Solang, 2000).

IV. CONCLUSION

1. Tilapia seeds measuring 3-5 cm can consume Apu-apu leaf meal meal. Where the feed with 20% Apu-apu leaf meal gave the highest response to the relative growth (168.99%) and the feed efficiency value (53.58%).
2. The use of Apu-apu leaf meal of more than 20% as in treatments E and B gave an unfavorable response to the relative growth and feed efficiency values.

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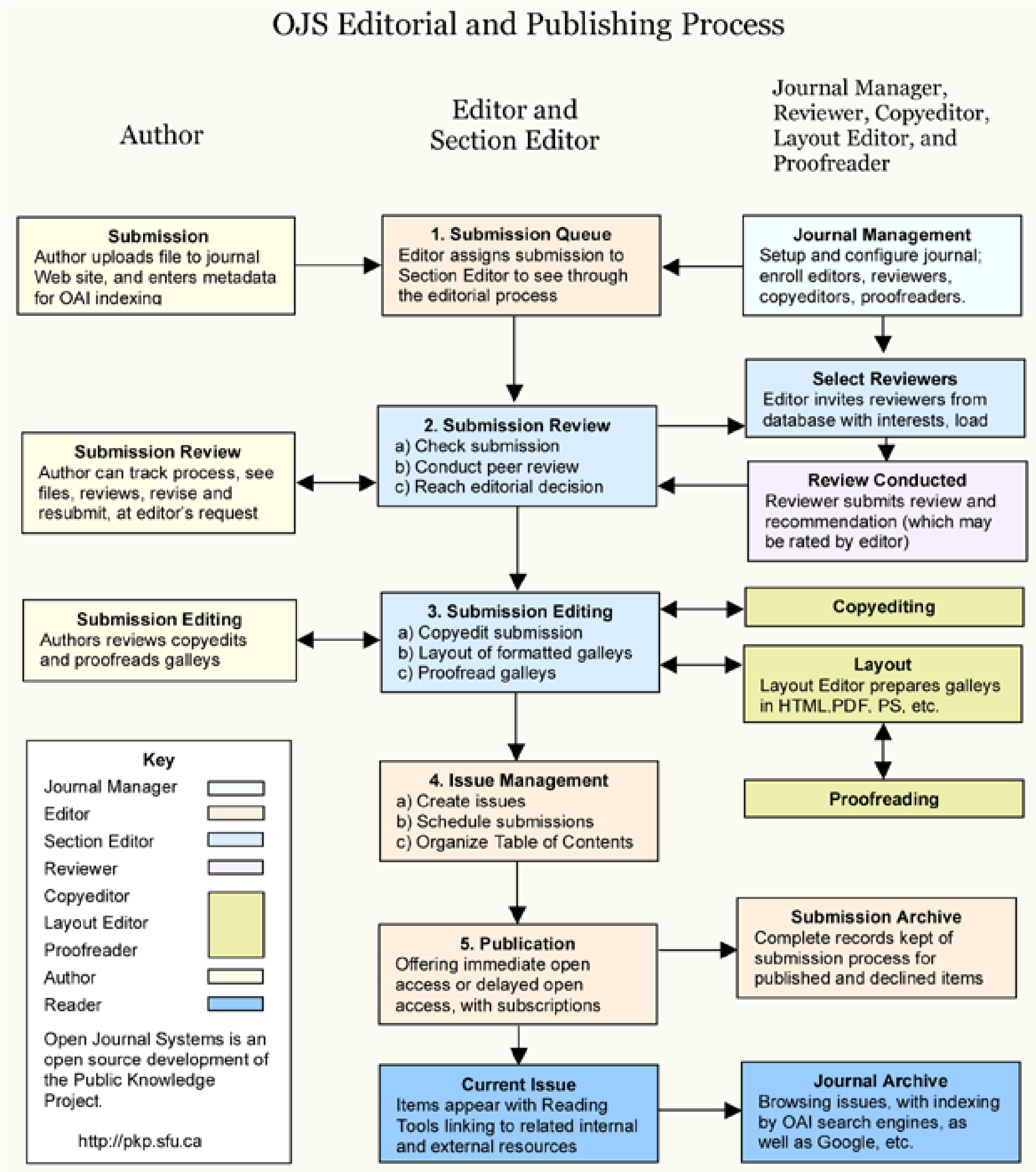

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