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Determinants of Post-Harvest Milk Losses among Milk Producers in Tanzania

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Abstract
Despite the Tanzania's programmes that aimed at improving infrastructures for milk and milk products, milk losses continue to be substantially high. The paper evaluated the determinants of post-harvest milk losses at the milk producers in Tanzania. The study adopted the cross-sectional research design whereby data were collected from 370 household heads and 38 Farm Managers in both the dry (June to October 2021; June to September 2022) and wet (November to December 2021; January to May 2022) seasons. Generally, study findings showed that milk was mainly lost through spillage, spoilage and contamination. In addition, Generalised Linear Mixed Model results showed existence of significant associations between location (Adjusted Coefficient (AC) = -0.80; 95% Confidence Interval (CI): -1.28-0.31), cattle keeping systems (AC = -0.75; 95% CI: -1.38-0.11), market price (AC = 0.66; 95% CI: 0.23-1.10), transport means (AC = -0.69; 95% CI: -1.29—0.10) and spillage. Additionally, washing cow's udder before milking (AC = -2.10; 95% CI: -3.70-0.49), water used for washing milk utensils (AC = -1.302; 95% CI: -2.43—0.17) and storage equipment (AC = -1.26; 95% CI: -2.47—0.05) were significantly associated with spoilage. Therefore, the Tanzanian government needs to improve extension services and workable strategies to minimise milk losses.
Keywords: Households, Milk Producers, Post-harvest Milk Losses, Tanzania

1.0. Introduction
The global population is projected to increase from 7.7 billion people in 2019 to 9.7 billion people in 2050 (UN, 2019). It is further projected population increase by 91% in sub-Saharan Africa (SSA) from 1.1 billion people in 2019 to 2.1 billion in 2050 (UN, 2019). Tanzania's population in 2022 was 61.9 million people and it is projected to increase by about 144% to 151.3 million in 2050 (URT, 2022b). The projected population growth signifies the need for collaborative efforts to increase production and minimise food losses to meet the increasing demand for food, dairy products included.

Milk losses are reported to be perpetuated by various factors in different areas of the world. According to Aulakh *et al.* (2013), handling and processing, production practices, management decisions, transportation facilities, infrastructure, consumer preferences and availability of financial markets are major factors associated with milk losses. It is also argued that a typical post-harvest chain comprises of a number of stages for the movement of harvested food from the site of production to the final retail market or consumer based on organisation and technologies. In addition, Aulakh *et al.* (2013) argue that in less developed

countries where the supply chain is less mechanized, larger losses are experienced during production handling, storage, processing and transportation.

Further to the above, Kowalska (2017) categorised the factors determining food losses and waste in two groups: first are the factors that occur along a food supply chain (FSC) e.g. machines (thus, investing in advanced technologies and new machinery is crucial for food losses and waste management especially in agricultural production and food processing); materials (quality of raw materials, develop thorough quality specification for acceptable materials); management (hence, need to develop and implement quality management systems); methods (proper technologies i.e. packaging technologies, processing technologies etc.); and people (knowledge, awareness and other attitudes of people engaged in a food supply chain). Second, are factors that come from the surroundings of a food supply chain e.g. political issues (policies, regulations, food and quality management standards, financial and substantive support for FSC operators 'public funds, preferential credit, professional training and education); consumer trends; consumer education; professional training; and food market development.

Furthermore, FAO (2019) identified various factors that drive food losses at supply chains i.e. on-farm level losses are caused by inadequate harvesting time, harvesting practices and handling, climate variability and product marketing. Others are inadequate storage conditions which impede products shelf life. In addition, during transportation, physical infrastructure and trade logistics play significant roles on preventing food losses. Furthermore, processing and packaging plays role in preserving foods, whereby, inadequate facilities as well as technical malfunction or human error can accelerate losses. At the retail level, food waste occurs due to limited shelf life and standard variations on demand. At consumption losses occurs due to poor purchase and meal planning, buying in excess, labelling misperception and poor storage at home (FAO, 2019).

Based on the above, Tanzania has formulated various policies, strategies and programmes aimed at contributing towards national food security through increased production, processing and by improving marketing infrastructure and marketing systems for livestock and livestock products (URT, 2006, 2010, 2011). In addition, the country insists on additional dairy investments and value addition through processing to ensure a stable market for fresh milk with the aim of reducing the losses along milk value chain (URT, 2017a). However, despite the government's initiatives, milk losses at production (household and farm levels) continue to be substantially high, approximately 6.5% (Lore *et al.*, 2005); 16% and 25% dry and rain seasons respectively (FAO, 2004 cited in ACF, 2014); 10% and 8% at the households and Farm levels respectively (Lugamara *et al.*, 2023 unpublished). Therefore, this paper evaluated the determinants of post-harvest milk losses at the milk producers (households and farms) node of Tanzania's milk value chain.

2.0. Materials and Methods

2.1. Description of Study Area

The study was conducted in three grouped livestock production systems of Tanzania namely: Central Zone (representing the agro-pastoral and semi-arid production systems); Coastal and Lake zones (representing the mixed rain-fed sub-humid and humid production systems); and the Northern and Southern Highland zones (representing a mixed rain-fed highland production systems) (Nell *et al*., 2014; URT, 2017a). In addition, about 30% and above of the households in the selected zones are engaged in livestock production (64% in the Central, 30% in Coastal and Lake; and 37% in the southern and northern highlands) (URT, 2017a).

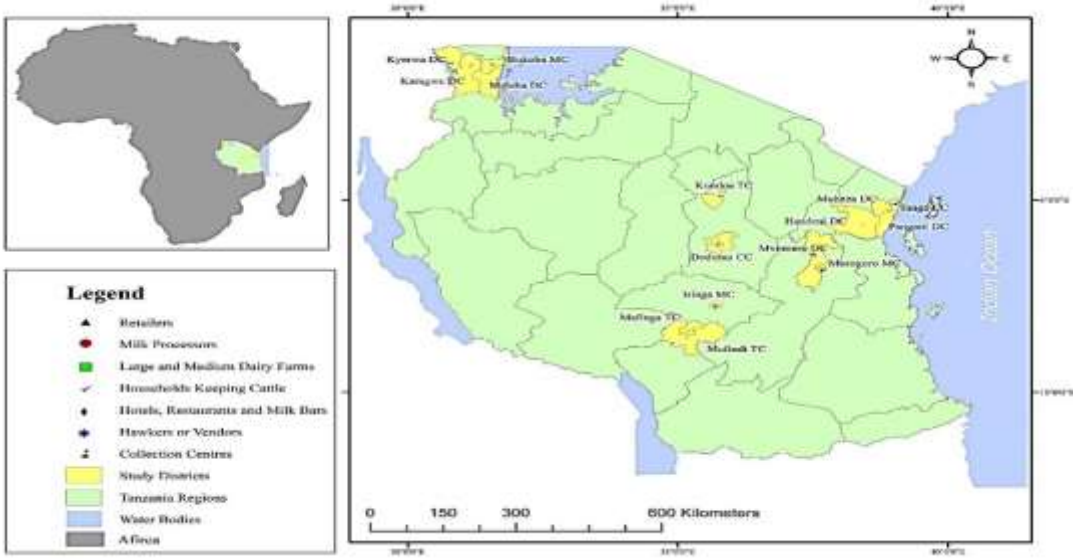

Figure 1: Map of Tanzania showing districts where the study was conducted

2.2. Research Design

A cross-sectional research design was used to collect data in both the dry (June to October 2021 in Morogoro, Dodoma and Tanga regions; June to September 2022 in Kagera and Iringa regions) and wet (November to December 2021 in Morogoro, Dodoma and Tanga regions; January to May 2022 in Morogoro, Dodoma, Tanga Iringa and Kagera regions) seasons. The research design is preferred because it allows determination of relationships between variables and can be done in a relatively short period of time while covering a large sample (Creswell, 2009; Gray, 2014; Kothari & Garg, 2014).

2.3. Sampling procedure and sample size

The study used a multistage sampling technique whereby livestock production systems covered are as explained in sub-section 2.1 above. In addition, five regions of Iringa, Morogoro, Kagera, Dodoma and Tanga were purposively selected from the sampled production systems. The regions were purposefully selected based on the number of household keeping cattle (NBS, 2017) and presence of cattle farms (TDB, 2019; URT, 2020). As part of sampling procedures, livestock production systems were considered as strata. Therefore, regions in each stratum were considered in the first stage, district in the second stage, wards third stage, villages fourth stage and households keeping cattle in the fifth stage. Further to the above, simple random sampling was used to select 370 household heads or their representative from households keeping cattle.

In determining the study's sample for households keeping cattle a random sample size calculation formula developed by Cochran (1977) was used as shown below:

$$n = Z^2_{\alpha/2} P\,(1 - P)/e^2 \quad \ldots\ldots\ldots\

household heads or their representatives because, 18 respondents (3.6% non-response rate) were dropped in the process (during the second round of interview their cows were no longer producing milk - dry off season). Moreover, at least 10% of farm managers representing dairy farms obtained from TDB and MLF reports and the regions or districts list were recruited for the study.

2.4. Data Collection methods

The study's quantitative data from milk producers was collected using two sets of questionnaires (i.e., one for the household and the other for the farm). The reason for using the different questionnaires was the differences in modes of operation and the nature of data to be gathered from the particular milk producers. The questions of each questionnaire were uploaded in a computer/mobile app (KoBoKollect) for easier and efficient data collection. The collected data was on main method of operation, individuals performing operation, equipment used, quantity handled, quantity lost and root causes of loss were recorded. Other information included cattle husbandry practices (systems used to keep cattle and procedures followed before milking), and respondents' socio-demographic and economic characteristics. The data were collected during or soon after milking to have the day's milk record. In addition, respondents were asked to report their milk loses in a one-month period as milk losses do not necessarily occur daily (daily based lose). In addition, key informant interviews (KIIs) were conducted to collect qualitative data. The key informants (KIs) included Regional and District Livestock and Fisheries Officers (RLFOs, DLFOs), Tanga Dairy Cooperative Union (TDCU), TDB (Tanzania Dairy Board), Tanzania Livestock Research Institute (TALIRI), African Dairy Genetic Gains (ADGG) – Tanzania, and Dairy Nourish Africa (DNA). The discussions with KIs mainly based on the main roots of milk losses and the factors associated with milk losses.

2.5. Data Analysis

The Statistical Package for Social Sciences (SPSS) software (version 26) was used to check the accuracy of data collected where anomalies found were corrected accordingly and performing descriptive analysis. The data were then transferred to STATA software for running Generalised Linear Mixed Model (GLMM) to determine the factors associated with milk losses at each stage of the milk node. The model was seen as desirable as is good in reducing the probability of having false positives based on the fact that the drivers of food losses are complex and interrelated (Grainger *et al.,* 2018). In addition, the likelihood ratio tests were undertaken to select the most suitable models. Furthermore, in order to overcome dependence problem resulting from repeated measures (data from two seasons) predictors (explanatory variables) were categorized into fixed and random variables. The random effect variables included villages for the households and farm identification number for the farms. Table 2-5 presents the Coefficients for the independent variables, the associated p-values and the 95% confidence interval (CI). Results are presented using adjusted Coefficient (Coef) and 95% CI. The differences/associations of variables were considered statistically significant if the *p*-value was ≤ 0.05.

3.0. Results and Discussion

3.1. Respondents Demographic and Socio-economic Characteristics

The results revealed that 83% of the households interviewed were headed by males and 17% were headed by females. This implies that a large number of households interviewed were headed by males compared to females. Also, head of the household had influences on access to and control of milk handling facilities and participation in milk operations hence leading to reducing post-harvest losses. A study by Zegeye and Teklehaymanot (2016) conducted in Ethiopia revealed that milking practices is mostly practised by men while milk handling, processing and marketing primarily practised by women. This may be due to the nature of activity involved especially hand milking which seems to be laborious in traditional cattle keeping. Other scholars (Alganesh, 2002; Tegegne *et al.,* 2013) reported women to perform many roles in milking and milk processing in some areas of Ethiopia while the role of men on milking and milk handling were not

significant. Therefore, understanding household characteristics particularly the head is of important factor when looking for post-harvest milk losses. In addition, the majority (65.1%) of household heads were in the age group of 36-60. Thus, suggesting the majority of household heads were in the economic active age group (URT, 2015), hence, able to participate in milk production. Further, the majority (91.4%) of respondents had formal education (i.e. seven years of primary school education and above) suggesting the household heads are in a position to understand proper livestock husbandry practices for better milk production and handling. Moreover, about three-fifths (59.5%) of the household heads are engaged in livestock production as their main economic activity. The findings suggest that, a household's economic status has an implication on the access of milking handling facilities for milk operations (during milking, collection, storage and transport). Therefore, strategies for minimising postharvest milk losses can have a substantial effect in uplifting the economic status of the households whose livelihood depends on livestock particularly, milk production. In addition, it is reported by FAO (2019) that, households' demographic characteristics i.e., age, education and sex of the household head, and household size need to be considered when looking for post-harvest food losses as associated factors.

Table 1: Demographic and Social economic characteristics of the household head (n = 370)

Variable	Category	Frequency	%
Sex	Male	307	83
	Female	63	17
Age	25-35	28	7.6
	36-60	241	65.1
	>60	101	27.3
Education level	None	32	8.6
	Primary education	163	44.1
	Secondary	77	20.8
	Tertiary (Certificate Diploma)	62	16.8
	University	36	9.7
Marital status	Single	7	1.9
	Married	299	80.8
	Divorced	11	3
	Separated	13	3.5
	Cohabitating	1	0.3
	Widow/er	39	10.5
Main occupation	Livestock production	220	59.5
	Crop production	61	16.5
	Government employees	37	10
	Private employees	21	5.7
	Self-employees and Casual labour (on and off farm)	31	8.4

Source: Field Data 2023

3.2. Milk losses at the Producers node

The study findings (Fig. 2) show that spillage, spoilage and contamination are the main causes of milk losses at the production node. According to Fig. 2, a total of 5.9%, 1.5%, and 0.4% of the milk was lost through spillage, spoilage and contamination in dry season respectively while the same caused 10.8%, 1.4%, 0.2% milk losses in the wet season respectively. At farm level the milk losses were approximated to a total of 4.4%, 2.2%, 0.1% by spillage, spoilage and contamination in dry season and 7%, 1.9%, 0.6% by spillage, spoilage and contamination in wet season respectively (Fig. 3). The above results signify the

existence of milk losses at the producers which impede the initiatives of fighting against poverty and hunger. The recorded milk losses are substantially high based on the milk consumption requirements and production costs per unit incurred. The study by Lore *et al.* (2005) conducted in Tanzania in 2003 reported 6.3% milk loss at farms (production area) due to spillage and spoilage. This implies that for many years the country (Tanzania) experiences substantial milk losses annually. Other scholars (Melesse *et al.*, 2014) reported milk losses at household level in Ethiopia to be 3.8% to 9.5% due to spoilage and spillage. Similarly, Ndungu *et al.* (2019) in Kenya and Zegeye and Teklehaymanot (2016) in Ethiopia reported milk losses at milk producers (smallholder dairy farmers) to be mainly of contamination, spoilage and spillage.

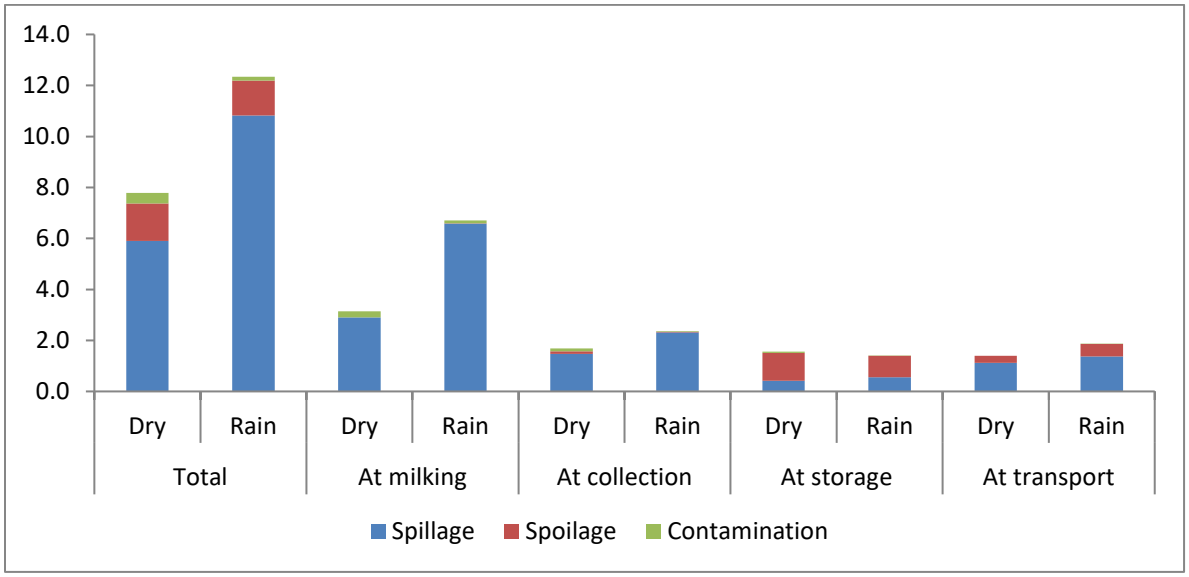

Figure 1: Percent of Milk losses by handling category at the household

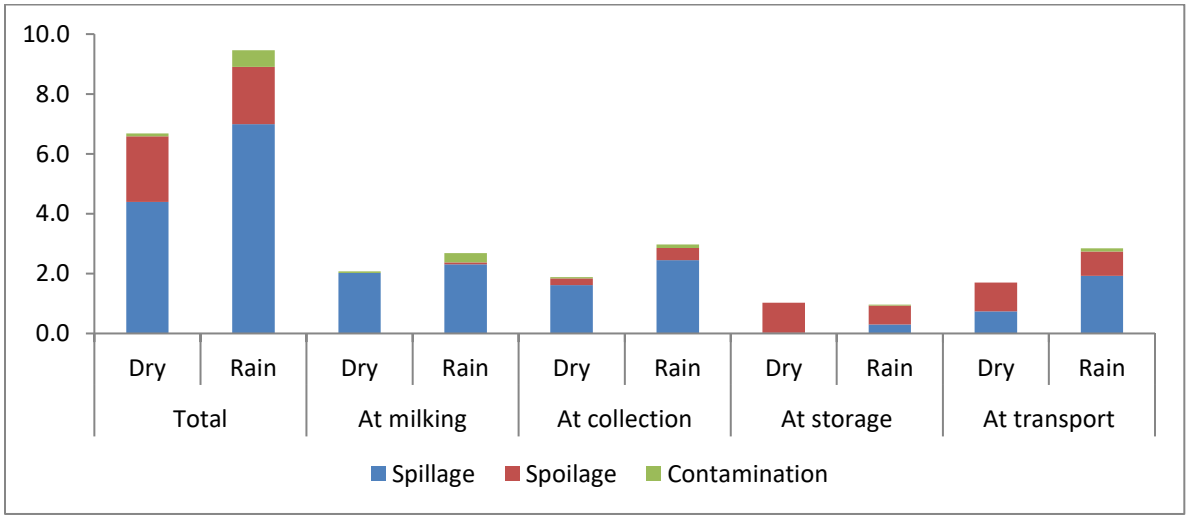

Figure 2: Percent of Milk losses by handling category at the farm

3.3. Determinants of post-harvest milk losses at the milk producers' node in Tanzania

The study assessed the determinants of post-harvest milk losses during milking, collection, storage and transport to market/sales point at the household and at the farm level. Since the outcome (dependent variable) is estimated milk losses in percentage terms and because of drivers for food losses being complex

and interrelated GLMM was used to assess the determinants (Grainger *et al*., 2018). The GLMM analysis results (Table 2) surprisingly showed that being married or cohabitating was significantly associated with PHML through spillage ($p<0.01$) during milking and collection ($p<0.05$). Table 2 also shows that household size was negatively and significantly ($p<0.01$) associated with milk losses by spillage during milking, collection, and transport to market. The results is contrary to the expectation that household with many members have more demand for food and other basic needs which are the basis for more commitment to minimise milk losses and ensure milk availability for the household's consumption and income.

Results in Table 2 further showed that an increase in a household's income was negatively and significantly associated with PHML through spillage ($p<0.01$) and spoilage ($p<0.05$) at collection. The observation seems to suggest that a unit increase in household income leads to milk losses. The plausible explanation may be most of the households in the area of study do not rely much on milk production for their livelihood rather than other sources of income. In addition, the results in Table 2 showed that extensive system for keeping cattle ($p<0.05$) and lack of proper milking area/place ($p<0.05$) were significantly associated with milk spillage. The above implies that the households using extensive system of keeping cattle experienced more milk losses compared to those using semi-intensive or intensive systems of keeping cattle because of lacking proper milking and collection facilities in the extensive system of keeping cattle. The higher losses also were on the households that did not use crush (not have proper milking place) for restraining cows during milking. The above may be due to the lack of knowledge and skills on milk handling practices, though some factors like whether respondent received training on milk handling practices, methods used in milking and whether respondent heard on PHML did not fit well when undertaking likelihood ratio tests to select the most suitable models for determining losses at milking and collection stages.

In addition, Table 2 showed a significant association between washing cow's udder before milking ($p<0.05$) and milk spoilage at storage. The above imply that households that were not washing the cow's udder before milking to minimise dirty experienced significant milk losses through spoilage. Table 2 further shows that households not using lubricants on cow's teat during milking experienced significant ($p<0.05$) milk losses through spillage during milking, this normally happens due to the fact that milking without lubricating the teats, the cow get restless and may feel pain and kick the milking container. Table 2 further shows that households not washing milk utensils was significantly associated with PHML through spoilage during storage ($p<0.01$) and during transport ($p<0.05$). Generally, households use of unsafe water to clean milk utensils was significantly ($p<0.05$) associated with milk losses by spoilage during storage than the households that used borehole/tap/rain water to wash milk utensils. The observation suggests that most of livestock keepers seem to use readily available water to wash the udder without considering its quality. Spillage also observed in the households that used jug, pot, calabash to handle milk when milking cows than the households that used bucket. The results conform to Ndungu *et al.* (2019) who reported milk spillage due to cow's knocking over milking equipment such as buckets during milking. Spoilage was observed at the households that used stainless steel containers to collect milk. This may be due to the delay at the collection to take milk to the storage or to be transported to market, because at normal circumstances stainless steel containers are advisable to use for milk collection. Also spoilage was significant to the households that stored milk without cooling facilities.

Table 2 also shows that households use of towels in drying udders during milking was positively and significantly ($p<0.05$) associated with PHML. Despite the fact that it is advisable to use towels to dry the cow's teats but, for this case it seems the towel was poorly used that's why milk spoilage and contamination were observed at the households claimed to use towel for drying cow's udders. The results in Table 2 showed a negative and significant association of being located in rural areas and postharvest milk losses by spillage ($p<0.01$) and contamination ($p<0.05$) during milk collection. The observation suggests that postharvest milk losses are less likely to occur in urban households compared to rural households.

Generally, in urban areas there are high possibilities of meeting the requirements needed for milking, milk collection, storage, transports, and markets and marketing compared to rural areas. The results above conform to what has been reported by Melesse *et al.* (2014) for Ethiopia that post-harvest milk losses vary between different geographical locations particularly rural and urban areas.

Table 2 also shows that PHML significantly ($p<0.01$ at milking and $p<0.05$ at collection) differed by season whereby, PHML were higher in the wet season compared to the dry season. In addition, there were more milk losses by spillage during this period. Furthermore, PHML due to contamination was also significantly ($p<0.01$) higher during the wet season at milking and collection. The cause for the above could be the fact that during the rainy season there were many milk outlets that outflowed the market and handling facilities hence losses by spillage. During the dry season milk supply is low compared to the demand which tempted milk producers to add some foreign materials like water (adulteration) and other substances to increase the volume of milk in order to get extra revenue while by doing so it led to milk contamination. The above results conform to what reported by FAO (2004) cited in ACF (2014) and NBS (2003) cited in Kurwijila *et al.* (2012) that milk losses is high in rain season compared to dry season. Similarly, Zegeye and Teklehaymanot (2016) and Kurwijila and Boki (2003) reported milk disposal because of adulteration. In addition the result is supported by Melesse *et al.* (2014) who reported severe problem of spillage during transfer of milk from container to container and during transportation process. Similarly, Amentae *et al.* (2015) conducted a case study in Ethiopia and found that poor milk handling practices, lack of appropriate milk handling facilities and milk mismanagement is associated with milk losses.

Table 2: Generalized Linear Mixed Model results on determinants of milk losses at the households

Dependent/ Independent Variables	Milking		Collection		
	Spillage	Contamination	Spillage	Spoilage	Contamination
Geographical location, 0 Rural, 1 Urban	-	-	-2.301 (-3.433--1.168) ***	-	-0.095 (-0.179--0.012) **
Season of survey, 0 Dry season, 1 wet season	3.641 (2.575-4.706) ***	-0.122 (-0.209-0.035)***	0.694 (0.050-1.339) **	-0.051 (-0.120-0.0172)	-0.099(-0.163-0.034) ***
Sex of Household head, 0 Male, 1 Female	-	-0.084 (-0.301-0.133)	-	0.091 (-0.120-0.0172)	-
Marital status of the Household head, 0 Otherwise, 1 Married/Cohabitating	2.755 (1.130-4.379) ***	-0.181 (-0.397-0.034) *	0.958 (0.027-1.889) **	-0.055 (-0.184-0.074)	-
Highest level of education of the Household head, 0 otherwise, 1 Secondary or above	1.678 (0.247-3.109) **	-0.111 (-0.228-0.005) *	-	-0.035 (0.104-0.033)	-
Household size, #	-0.7419 (-1.143--0.340) ***	-0.009 (-0.042-0.024)	-0.286 (-0.507--0.064) ***	-	-
Annual household income, Tzs	-2.27e-08 (-5.66e-08-1.11e-08)	-2.24e-09 (-5.02e-09-5.44e-10)	-2.48e-08 (-4.35e-08--6.11e-09) ***	-1.78e-09 (-3.43e-09--1.37e-10) **	-
System for keeping cattle, 0 Otherwise, 1 Semi/Intensive	-	0.148 (-0.032-0.328)	-	-	-
Place of milking cow, 0 Otherwise, 1 In crush	-2.098 (-3.778--0.417) **	-	-0.880 (-1.829-0.069) *	-0.078 (-0.163-0.008) *	-
Material used for drying cows teats, 0 Otherwise, 1 Towel	-	-	-	0.055 (-0.025-0.134)	-

Whether use lubricant on teats during milking, 0 No, 1 Yes	-	-0.168 (-0.314-0.023) **	-	-	
Type of Lubricant used on teats during milking, 0 Otherwise, 1 Milk salve	-	-	-	-	-0.036 (-0.116-0.044)
Whether wash milking utensils, 0 No, 1 Yes	-	-0.158 (-0.433-0.116)	-	-0.060 (-0.214-0.094)	-
Type of water used to wash milking utensils, 0 Otherwise, 1 Borehole/tape/rain	-	-	-	-0.069 (-0.151-0.014) *	-
Whether use detergent to wash milking utensils, 0 No, 1 Yes	-	0.166 (0.031-0.301) **	-	-0.052 (-0.125-0.021)	-
Method of milking, 0 hand milking, 1 Machine milking	-	-1.367 (-1.86--0.874) ***	-	-	-
Equipment used during milking, 0 Otherwise, 1 Bucket (plastic/iron)	-10.819 (-14.28--7.357) ***	-	-	-	-0.228 (-0.404--0.052) **
Equipment used for milk collection, 0 Otherwise, 1 aluminium/ stainless steel Milk can	-	-	-	0.166 (0.045-0.287) ***	-
The person performing milk collection, 0 Hired labour, 1 Family members	-		-0.600 (-1.558-0.158)	0.046 (-0.027-0.119)	
Adequacy of labour for milk operations, 0 No, 1 Yes	-	-	-	-	0.102 (-0.033-0.236)
Market availability for milk sales, 0 No, 1 Yes	-	-	1.192 (0.0193-2.365) **	-	
Membership milk association, 0 No, 1 Yes	-1.9148 (-3.967-0 .139) *	-	-1.816 (-3.028--0.605)***	-	-
Market availability, 0 Otherwise, 1 Available	-	-	-	-0.063 (-0.138-0.011) *	
Market distance, 0 long distance (>6km), 1 short distance(≤6 km)	-	-	-	-0.152 (-0.233--.071) ***	
Constant	14.45 (10.395-18.506) ***	0.517 (0.13-0.903) ***	3.461 (1.482-5.441) ***	0.572 (0.313-0.832) ***	0.312 (0.106-0.518) ***
Number of observations	740	740	653	653	740
Log likelihood	-2583.154	-724.229	-1883.225	-307.293	-464.405
Chi2	123.77	76.13	64.96	62.45	26.75
P	0.000	0.000	0.000	0.000	0.000
Random effects					
Village var(_cons)	17.519 (11.014-27.867)	0.109 (0.071-0.168)	3.944 (2.522-6.167)	0.007 (003-0.017)	0.006 (0.002-0.020)
Village var(dependent)	54.591 (48.897-60.949)	0.361 (0.324-0.402)	16.492 (14.697-18.505)	0.144 (0.129-0.161)	0.201 (0.180-0.223)
Chi2	76.02	97.46	83.32	13.31	5.15
P	0.000	0.000	0.000	0.001	0.0116

NB: Number outside the bracket refers to adjusted coefficient while the number in bracket indicates 95% confidence interval. ***, **, * are significance levels at 1%, 5%, and 10%, respectively

Results further shows that personal characteristics were associated with PHML for example, PHML were relatively higher when family members participated in transporting the milk compared to when done by hired labour: the association was statistically significant ($p<0.001$) (Table 3). The observation seems to suggest that some household members were either non-experienced or negligent in handling milk hence the losses. In addition, the results in Table 3 showed significant association between being located in rural areas ($p<0.001$), large household size ($p<0.01$), extensive system of keeping cattle ($p<0.05$), knowledge on

PHML (0.01), lack of refrigeration facilities for milk cooling ($p<0.05$), poor road to market ($p<0.05$) and milk losses by spillage during storage and transport to market. Moreover, milk spoilage during storage and transport to market at the households were significantly associated failure to wash cow's udder before milking ($p<0.05$), improper use of towel to dry cow's teats ($p<0.05$), improper cleanness of milk utensils or use of unclean water to wash milk utensils ($p<0.05$), use of storage equipment with no cooling facilities ($p<0.05$) and poor road to market ($p<0.05$). Likewise, bad use of disinfectant before and after milking ($p<0.01$) was significantly associated with milk contamination. The study results (Table 3) revealed a positive and significant association between awareness or knowledge of the household head on post-harvest milk losses and milk spillage ($p<0.01$) and spoilage ($p<0.05$) particularly when transporting milk to market. This implies that knowledge and skills are not a panacea to PHML, unless other factors like market availability, accessibility and means of transport including good roads which are passable all the time to market are taken into consideration. Except the knowledge and skills which shown a positive influence on PHML, the results in Table 3 conform to Zegeye and Teklehaymanot (2016) who reported poor milk handling practices, contamination, lack of cooling facilities, long distance to market, use of inappropriate containers, lack of market and delays of transport to have influences on milk losses. In addition, Amentae *et al.* (2015) found lack of cooling systems at the household and during transport; poor means of transportation; inappropriate milk carrying equipment and poor storage facilities to be associated with milk losses. Similarly, Lore *et al.* (2005) reported spillage and spoilage at transportation caused by the use of inappropriate containers to transport milk, poor hygiene, low level of technology application for milk preservation to be the factors influencing milk losses in Ethiopia and lack of markets and poor roads in Tanzania.

Table 3: Generalized Linear Mixed Model results on determinants of milk losses at the household

Dependent/ Independent Variables	**Storage**			**Transport**		
	Spillage	**Spoilage**	**Contamination**	**Spillage**	**Spoilage**	**Contamination**
Geographical location, 0 Rural, 1 Urban	-0.797 (-1.281-0.312) ***	-	-0.034 (-0.091-0.023)	- 1.773 (-3.562-0.0154) *	-0.314 (-0.773-0.145)	-
Season of survey, 0 Dry season, 1 Rain/wet season	-	-	-0.029(-0.085-0.0271)	-	0.200 (-0.116-0.517)	-
Sex of Household head, 0 Male, 1 Female	0.409 (-0.062-0.880) *	-	-	-		-
Marital status of the Household head, 0 Otherwise, 1 Married/Cohabitating	-	-	-	0.726 (-0.399-1.851)		-
Highest level of education of the Household head, 0 otherwise, 1 Secondary or above	-0.156 (-0.551-0.239)	-	-0.048 (-0.106-0.010)	-		-
Household size, #	-0.065 (-0.179-0.048)	-	-	-0.456(-0.739--0.172) ***	-0.046 (-0.152-0.060)	-
Annual household income, Tzs	-	-	-	-8.85e-09(-2.87e-08-1.10e-08)	-5.38e-09(-1.50e-08-4.22e-09)	-
System for keeping cattle, 0 Otherwise, 1 Semi/Intensive	-0.7476 (-1.384-0.112) **	-	-	-	-	-
Place of milking cow, 0 Otherwise, 1 In crush	-0.176 (-0.634-0.282)	-	-	-	-	0.005 (-0.003-0.013)
Whether wash cow udder before milking, 0 No, 1 Yes	-	-2.095 (-3.704-0.486) **	-	-	-	-
Material used for drying cows teats, 0 Otherwise, 1 Towel	-	1.023 (0.113-1.933) **	-	-	-	-

Whether use disinfectant before and after milking, 0 No, 1 Yes	-	-	-	-	-	0.077 (0.060-0.093) ***
Whether wash milking utensils, 0 No, 1 Yes	-	-3.785 (-6.014--1.556)	-	-	-0.893 (-1.771--.01489) **	-
Type of water used to wash milking utensils, 0 Otherwise, 1 Borehole/tape/rain	-	-1.302 (-2.430--0.174) **	-	-	-0.359 (-0.834-0.116)	-
Equipment used milk storage, 0 Otherwise, 1 With cooling facilities	-	-1.2628(-2.471--0.054) **	-	-	-	-
The person performing milk transport, 0 Hired labour, 1 Family members				0.864 (-0.082-1.809) *		0.012 (0.005-0.019)
Adequacy of labour for milk operations, 0 No, 1 Yes	0.400 (-0.338-1.137)	-	-	-	-	-
Market availability for milk sales, 0 No, 1 Yes	-	1.595 (0.219-2.971) **	-	-	-0.420 (-0.978-0.138)	-
Whether selling milk at home, 0 No, 1 Yes	-	-	-	-	-	-
Membership milk association, 0 No, 1 Yes	-0.159 (-0.756-0.438)	-	-	-	-	-
Whether heard on postharvest milk losses, 0 No, 1 Yes	-	-	-	2.877 (0.769-4.985) ***	0.742** (0.047-1.436)	-
Adequacy of milk storage facilities, 0 Otherwise, 1 Adequate	0.198 (-0.213-0.608)	0.457(-0.54-1.454)	0.048 (-0.009-0.105) *	-	-	-
Availability of refrigeration facilities, 0 Not available, 1 Available	-0.528 (-1.053--0.003) **	-				
Electricity for milk cooling, 0 Not Stable/Not electricity, 1 Stable		-0.836 (-1.988-0.315)				
Market availability, 0 Otherwise, 1 Available	-	-	0.030(-0.037-0.097)	-	0.338 (-0.092-0.768)	-
Market distance, 0 long distance (>6km), 1 short distance (≤6 km)	-	-0.253 (-1.141-0.635)	-0.023 (-0.081-0.035)	-	-	-0.007(-0.014--0.001) **
Milk market price, 0 Low price, 1 Good/reasonable price	0.661 (0.226-1.096) ***	0.499 (-0.554-1.551)	0.039 (-0.026-0.104)	0.771 (-0.504-2.046)	-	-
Transport means to market using road, 0 Poor road, 1 Good and passable all the time	-0.694 (-1.291--0.097) **	-2.771(-4.300--1.243) ***	-	-0.861 (-2.381-0.660)	-	-0.012(-0.022--0.001) **
Constant	2.089 (0.918-3.260) ***	10.044(6.055-14.033) ***	0.062(-0.056-0.181)	3.365 (0.762-5.968) **	1.869 (0.840-2.898) ***	0.0138(-0.001-0.029) *
Number of observations	687	740	740	592	740	592
Log likelihood	-1521.344	-2314.635	-323.766	-1755.743	-1644.844	1115.975
Chi2	57.82	63.72	15.2	29.73	28.91	130.98
P	0.000	0.000	0.034	0.000	0.000	0.000
Random effects						
Village var(_cons)	0.340 (0.224-0.712)	3.063 (1.529-6.137)	-	16.124 (10.86383-23.932)	0.229 (0.086-0.612)	0.0001 (0.0001-0.0003)
Village var(dependent)	4.620 (4.141-5.153)	28.445 (25.523-31.701)	-	16.680 (14.683-18.948)	4.809 (4.323-5.349)	0.001 (0.001-0.001)
Chi2	40.81	15.41	-	126.66	6.52	36.88
P	0.000	0.000	-	0.000	0.038	0.000

NB: Number outside the bracket refers to adjusted coefficient while the number in bracket indicates 95% confidence interval. ***, **, * are significance levels at 1%, 5%, and 10%, respectively

Source: Field data 2023

Further to the above Table 4 shows milk losses at the farm level. The most observed determinants were season of survey whereby rain season shown positive and significant influence on milk losses by contamination ($p<0.05$). In addition, maximum distances to market shown positive and significant influences on milk losses by contamination ($p<0.01$) (Table 5).

Table 4: Generalised Linear Mixed Model results on determinants of milk losses at the farm

Dependent/ Independent Variables	**Milking**			**Collection**
	Spillage	**Spoilage**	**Contamination**	**Spoilage**
V Geographical location, 0 Rural, 1 Urban	-	-	0.157 (-0.071-0.385)	-
Season of survey, 0 Dry season, 1 Rain/wet season	-	-	0.269 (0.048-0.489) **	-
Sex of the farm manager, 0 Male, 1 Female	-	-	0.292 (-0.163-0.746)	-
V Grazing System, 0 Otherwise, 1 Semi/Intensive	-0.916 (-2.211-0.378)	-0.142 (-0.285-0.0006) *	-	-
Place used to milk cow, 0 Otherwise, 1 in crush	-	-	-	-0.502 (-1.199-0.194)
Type of lubricant used, 0 Otherwise, 1 Milk salve/Norbrook	-	-	-	-0.566 (-1.278-0.147)
Water used to wash milk utensils, 0 Otherwise, 1 Borehole/tape/ rain water	-	-	-	0.446 (-.196-1.089)
Whether use detergent to wash milking utensils, 0 No, 1 Yes	-	0.17 (.009-0.331)	-	-
Whether heard on postharvest milk loss, 0 No, 1 Yes	-1.063 (-2.505-0.379)	-	-	-
Constant	3.010 (1.958-4.063) ***	0.075 (-0.034-0.183)	-0.040 (-0.223-0.143)	0.88 (0.095-1.66) **
Number of observations	76	76	76	76
Log likelihood	-184.871	-12.351	-53.607	-132.863
Chi2	4.59	6.09	9.68	8.59
P	0.101	0.048	0.022	0.035
Random effects				
Farm ID var(_cons)	-	0.003 (8.38e-08-84.853)	-	-
Farm ID var(dependent)	-	0.078 (0-0.125)	-	-
Chi2	-	0.04	-	-
P	-	0.425	-	-

Table 5: Generalised Linear Mixed Model results on determinants of milk losses at the farm

Dependent/ Independent Variables	**Storage**			**Transport**	
	Spillage	**Spoilage**	**Contamination**	**Spillage**	**Spoilage**
V Geographical location, 0 Rural, 1 Urban	-0.235 (-0.630-0.160)	-	-	-	-
Season of survey, 0 Dry season, 1 Rain/wet season	0.267 (-0.118-0.652)	-	-	1.185 (-0.408-2.775)	-
Frequency of milking per day, 0 Otherwise, 1 Twice per day (morning and evening)	-0.350 (-0.796-0.097)	-	-	-	-
Water used to wash milk utensils, 0 Otherwise, 1 Borehole/tape/ rain water	-	-	-	-	0.533 (-0.339-1.405)
Method of milking, 0 hand milking, 1 machine milking	-	1.296 (-0.387-2.979)	-	-	-
Selling milk at home/farm, 0 No, 1 Yes	-	0.915 (-0.0382-1.868) *	0.013 (-0.040-0.067)	-	-
Place for selling milk, 0 Otherwise, 1 MCC/Processors/Cooperatives	-	-	-	-	0.640 (-0.248-1.528)
Maximum distance to market, kilometres	-	-	0.002 (0.001-0.002) ***	-	-

Whether received training milk handling, 0 No, 1 Yes	-0.274 (-0.695-0.146)	-	-	-1.155 (-2.767-0.457)	-
Adequacy of milk storage facilities	-	-0.495 (-1.466-0.476)	0.014 (-0.052-0.080)	-	-
Availability refrigeration facilities, 0 Not available, 1 Available	-	-	-0.015 (-0.087-0.058)	-	-
Market distance, 0 long distance >6km, 1 Short distance ≤6km	-	-	-	-	-0.284 (-1.179-0.610)
Market Price, 0 low price, 1 Good/reasonable price	-	0.855 (-0.030-1.740) *	-	-	-
Transport to market using road, 0 Poor road, 1 Good and passable all the time	-	-	-	-	0.649 (-0.458-1.755)
Means of transport to market, 0 Otherwise, 1 By tri/motorcycle/car	-	-	-	-	0.6876 (-0.461-1.856)
Constant	0.523 (0.088-0.957) **	-0.248 (-1.306-0.81)	-0.022 (-0.068-0.023)	1.411 (-.051-2.874) *	_-0.047 (-1.349-1.256)
Number of observations	76	76	76	76	76
Log likelihood	-95.986	-148.490	69.539	-203.927	-153.94284
Chi2	10.25	10.12	70.45	4.1	9.39
P	0.037	0.039	0.000	0.129	0.1529
Random effects					
Farm ID var(_cons)	-	0.816 (0.240-2.769)		-	-
Farm ID var(dependent)	-	2.211 (1.410-3.467)		-	-
Chi2	-	2.86		-	-
P	-	0.0453		-	-

4.0. Conclusion and Recommendation

Reducing milk losses at production level is among the most impactful strategies to meet the optimal food and nutrition security of the growing population globally. The study evaluated determinants of post-harvest milk losses at the milk producers (households and farms), particularly the main forms of milk losses at the milk producers and the determinants that perpetuate milk losses in the area of study. Generally, study findings show that spillage, spoilage and contamination were the main forms of milk losses at each stage of milk handling especially during milking, collection, storage and transport to market. In addition, GLMM results showed that husbandry practices, socio-economic factors, geographical locations, season of production were positively and significantly associated with milk losses. In addition, poor milk handling facilities, storage without cooling facilities, long distance to market, poor road and market price were significantly associated with post-harvest milk losses at the households. Furthermore, poor membership to milk association and lack of education on post-harvest milk losses were significantly associated with post-harvest milk losses. Therefore, the Tanzanian government should improve extension services to impact knowledge to milk producers on milk handling practices including use of proper milk equipment during milking, collection, storage and transport, and better husbandry practices so as to minimize PHML. In addition, the government should come up with workable strategies or actions to minimise post-harvest milk losses by milk producers in order to increase their returns and save Tanzania from relying on milk imports that drain the country's foreign currency.

Limitation of the study and areas for further research

The study results are based on the respondent's self-reported postharvest milk losses occurring at each node of the value chain. Despite the limitation, the researchers carefully designed the questionnaire and the data collection exercise was carried out by well-trained enumerators. Nonetheless, further research is needed to consider a larger sample size and include other regions in Tanzania to provide a more comprehensive picture of postharvest milk losses in the country.

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Nutrition-Sensitive Agriculture and Appropriate Postharvest Handling of Crops: A Missing Link with Potential for Global Nutrition and Food Security

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Abstract
Currently, 822 million, and 2 billion suffer from hunger, and micronutrient deficiency, overweight and obesity, respectively. This is due to a variety of factors associated with pre-harvest and post-harvest activities in nutrition-sensitive agriculture. This paper is, therefore, aimed at reviewing nutrition-sensitive agriculture and appropriate postharvest handling of crops: a missing link with potential to global nutrition and food security. Agriculture provides food and nutrients through improved agricultural production systems such as biofortified and/or crop breeding, crop diversification, fish, poultry, and animal rearing, use of biodiversity, and urban and peri-urban agriculture. Appropriate postharvest handling such as right harvesting time, methods and maturity stage, field packing, transportation, storage, processing, packing, safety and distribution of the products help to safeguard the quality of the product and reduce postharvest food wastage. However, the agriculture and postharvest system of most crops in developing countries are constrained by lack of a complete package of agricultural inputs, income, facilities/technologies, tradition and religion, and lack of awareness on how to use technologies and protect the safety and or quality of the products. The interventions that make agriculture and the postharvest system of crops nutrition-sensitive should be appropriately applied to the crop and postharvest system of crops.
Keywords: Agriculture, Food security, Nutrition security, Nutrition-Sensitive, Postharvest handling.

1. Introduction

Studies indicate that about 822 million are food insecure and 2 billion people have micronutrient deficiency and are overweight or obese (FAO, 2022). The developing countries account for 91% of stunted children worldwide, with Sub-Saharan Africa accounting for more than one-third (UNICEF/WHO/WorldBank, 2021). To reduce the number of vulnerabilities, the United Nations modified the Millennium Development Goal in 2015 to the Sustainable Development Goal, which states that by 2030, 'end hunger, achieve food security, improve nutrition, and promote sustainable agriculture' will be achieved (SDG, 2016).

Food security exists when all people, at all times, have physical and economic access to sufficient, safe, nutritious food to meet their dietary needs and food preferences for an active and healthy life". It is most commonly defined as the four pillars: availability, access, utilization, and stability (Peng and Berry, 2019). In other words, nutrition security exists when food security is combined with a sanitary environment, adequate health services, and proper care and feeding practices to ensure a healthy life for all household members (FAO, 2012). Political insecurity, war, macroeconomic inconsistencies, trade displacement to ecological deprivation, poverty, population growth, gender disparity in food production, illiteracy, and inadequate health services are all factors that contribute to food insecurity (Birara et al., 2015).

Agriculture provides food and earnings for the human beings necessitated to survive.It is, however, only concerned with increasing output and productivity and cannot effectively contribute to nutrition (Chihambakwe et al., 2019).

Poor agricultural production and handling practices, on the other hand, result in postharvest loss and food insecurity. Consequently, food insecurity leads to malnutrition, which negatively affects agricultural productivity, socioeconomic development, educational performance, and community health (CWFC, 2012). Farmers with poor nutritional status have lower agricultural productivity, household income, and health. Thus, interventions that make agriculture more nutrition-sensitive are critical (de la Pena et al., 2018).

Nutrition-sensitive agriculture is a food-based approach to agricultural development that places nutritionally rich foods, dietary diversity, and food fortification at the heart of overcoming malnutrition. Making agriculture a nutrition-sensitive is an approach that seeks to ensure the continuous production of a variety of low-cost, adequate nutrient-dense, locally acceptable, and hazard-free foods in sufficient quantities and quality to meet the needs of consumers (Uccello et al., 2017). It also requires a good agricultural policy on food prices and consumption, women's employment in agriculture, and child feeding, nutritional status, and health conditions (Gillespie and van den Bold, 2017).

Though agriculture's primary role in nutrition improvement is to improve access to sufficient, safe, and nutritious foods, the expected benefit is not realized due to inadequate agricultural inputs, biotic and abiotic stresses, a poor social and infrastructural situation, a weak extension service, poor governmental regulations and legislations, labor wastages, a chronic shortage of cash, and poor women empowerment (de la Pena et al., 2018). Even though it is advised that food and nutrition security be ensured through nutrition-sensitive agriculture and proper crop postharvest handling, it can be challenging to find reliable, organized information. Therefore, this paper aims to review nutrition-sensitive agriculture and appropriate postharvest handling of crops: a missing link with potential for global nutrition and food security.

2. Nutrition-sensitive agriculture

2.1. Agriculture as a source of income

Agriculture affects nutrition by influencing money spent on purchasing a variety of nutritious food; access to a consistent source of income; and the role of women in agriculture (Dennison, 2014). Ensuring consumers' regular and adequate income is necessary to attain nutritious food, access to health care and education services. This is accomplished by increasing the potential of agriculture and food systems to generate income and increasing the likelihood that the income is used on purchasing nutritious foods and accessing nutrition-enhancing services (Uccello et al., 2017).

2.2. Agriculture as a source of food

The principal purpose of agriculture is not to provide the food types required by families, as most rural low-income families are net purchasers of food (AFC, 2018). Food security for families with access to arable land is determined by the food produced for consumption, income, and local food availability and prices, while the type, quantity, and seasonality of food available in the household for consumption are affected by food production (Chihambakwe et al., 2019). The availability and diversity of food in local marketsare also influenced by food production (Herforth and Harris, 2014).

The role of agriculture as a source of nutritious food is indicated through several interventions reported by several researchers. For instance, the availability and consumption of microelement-rich foods reduced microelement malnutrition in Bangladesh, South Africa, and Ethiopia through home gardening connected to primary health care activity and nutrition (Talukder et al., 2000). Furthermore, own production improves family nutrition because they use their products at home (Chihambakwe et al., 2019). Similarly, the production of diversified farming by crop-livestock incorporation with market accessibility improved the nutrition of family members in Zimbabwe (Murendo et al., 2018). Crop diversification, cattle ownership, credit access, and education also improved Mali's food consumption score (Mango et al., 2018).

Livestock and animal-based foods are critical to long-term sustainability. They play an important role in improving nutrition, reducing poverty, promoting gender equity, enhancing livelihoods, increasing food security, and strengthening human health (Adesogan et al., 2020). Animal source food (ASF) is a good source of protein, as well as certain micronutrients (easily absorbable iron, zinc, calcium, vitamin A, vitamin B12, and various essential amino acids) (Neumann et al., 2014). The link between animal-source food consumption and stunting reduction is also reported, indicating that food supplementation with a small amount of milk and meat improves macro and micronutrients dramatically.Moreover, meat supplementation also improves cognitive skills, leadership behavior,

physical activity, and initiative (Headey et al., 2018). Similarly, fish-based foods are high in omega-3 fatty acids and iodine, which are essential for children's brain development (Thilsted et al., 2016). However, consumption of animal-source foods is subjected to religious custom and practices, which excluded women and children from some foods. Because of this, most foodstuffs were not permitted for pregnant women. Pregnant women, for example, were not permitted to consume meat and fish products, potatoes, fruits, beans, eggs, butternut, or pumpkin (McNamara and Wood, 2019).

Biodiversity is also viewed as a means of achieving dietary diversity and ensuring nutrient sufficiency. For instance, an increase in species diversification resulted in a 12.7% improvement in micronutrient adequacy in Kenya (Oduor et al., 2019). Similarly, households with higher agricultural biodiversity scored higher on dietary diversity and had greater food security than those with lower agricultural biodiversity (M'Kaibi et al., 2017). Moreover, the use of underutilized food crops such as *Dolichos (*Minde et al., 2021), Amaranth (Aderibigbe et al., 2020), and Anchote (Mosissa et al., 2019), African native fruit plants (Omotayo and Aremu, 2020) as well as the fortification of neglected legume and oilseed crops with cereals (Desire et al., 2021), and use of local agro-forestry practices Jemal et al., 2018), have improved the nutrition. Some underutilized wild animals, such as rats,were also reported to be good protein sources (Broegaard et al., 2017).

In Sub-Saharan African countries, breeding programs have also improved nutrient density, for example, maize is made suitable for local processing of bread and snacks, the yield has increased, a loss is reduced, and anti-nutritional factors are reduced (Ekpa et al., 2018). The urban and peri-urban agriculture (UPA) program also enabled residents to grow crops, medicinal herbs, fruits, raise small animals, and participate in an integrated farming system, which improves their food and nutrition security.This UPA enables low-income urban residents to obtain fresh, diverse, and nutritious food near the market(Nisbett et al., 2017). The food producers in urban areas benefit in two ways: their food requirement is fulfilled, and the expenses incurred for food are reduced (Chihambakwe et al., 2019). Since there are no more agricultural lands in urban and peri-urban areas, intensification of farming is very critical to encourage agricultural production to promote food and nutrition security (Smith, 2013).

3. The appropriate postharvest handling of crops in nutrition-sensitive agriculture

3.1.Appropriate postharvest handling practices of perishable crops

3.1.1.Harvesting

Postharvest handling begins at harvest. Nonetheless, the product's postharvest quality is determined by pre-harvest factors such as environmental (relative humidity, temperature, light, rains, and winds) and cultural factors (nutrient management, cultivar, disease management, irrigation, agronomic practices, and economy) (Weber, 2020).Improper harvest, maturity stage, harvesting time, and methods are causing losses on crops after harvest (Kumar and Kalita, 2017). The majority of the losses of fruits at the farmer’s level occur during storage due to the harvesting of immature fruits or the fruit being overripe (Omotayoand Aremu, 2020). Therefore, during harvesting, mechanical damage (cuts, scratches, and abrasions) should be minimized because it affects the appearance of the perishable crops and is a pathway for microorganisms that deteriorate at transportation and storage (Kuiper et al., 2020). Harvesting at the propermaturity stage also helps attain fruit quality and reducesthe chance of spoilage (Mutungi et al., 2020).

3.1.2.Packing or Field handling

Packing is one of the activities performed to place detached produce into containers used for transportation. Before packing the produces, remove extraneous materials, wash to remove soil and microbial load, pre-cool to reduce heat physiological activities, sort based on required size categories, inspect the quality of the produces, and pack into the appropriate containers during the transportation is very important to reduce the postharvest loss (Alistair, 2005). To reduce the loss in the packing house, careful transfer of produce from containers used to pick to field crates is necessary as it reduces mechanical damage. Similarly, soil-borne microorganisms should be protected by avoiding direct soil contact withthe crop. It is also critical to place containers in a shaded area and fill them to the recommended filling weight (Behera et al., 2020).

3.1.3.Transportation

The primary transportation step is to hold the crop from the productionarea to the selling place by truck, rail, ship, plane, pickup truck, and human back, animals (donkey, horse, or mule). Unlike developed countries, the transportation system is constrained by the lack of a cold transportation system and does not haveventilation facilities to transport perishables in developing countries (Arah et al., 2016). In Ethiopia, for instance, all transportation methods predispose

perishable produce to heat buildup and mechanical damages due to impact force, vibration, and damage due to humans loading over the crop and improper and inadequate roads (Kuyu and Tola, 2018). Furthermore, delays in transportation without the use of cushioning materials, uncovering the vehicle, transporting during the hot time of day, mixing the products, overfilling, loading with other harmful chemicals, and a lack of refrigerated transport were reported as constraints in developing countries (Emana et al., 2017; Kuyu and Tola, 2018).

3.1.4. Storage

Food storage preserves the quality of the products for an extended period until consumption. Good storage facilities help to extend the shelf life of produce and ensure a sustainable supply of the product throughout the value chain (Arahet al., 2016). As a result, the produce season of availability is prolonged, and the product can be kept till prices rise, creating a uniform retail distribution. It has been demonstrated that products stored in refrigerated storage, controlled atmosphere, modified atmosphere packaging, and hypobaric storage has a longer shelf life (Kaur et al., 2021).The commodities stored together should be checked for their initial quality, stability, environmental and handling practices, compatibility, optimum temperature, relative humidity, and other environmental factors (Lee and Kader, 2000). Before storage, produce should be sorted into healthy and unhealthy categories, and the maturity stage of the product should be considered, with high-quality products stored together (Arah et al., 2016). If different crops are to be stored together, the compatibility of the produces should also be checked first (Table 1). Grain products should also be dried to reduce their high moisture content before being stored to prevent a microbial attacks and insect infestation(Asemu et al., 2020). The storage facilities, warehouse, and environment must be protected from all kinds of environmental hazards. Grain storage in steel bins and plastic silos is preferable because it provides the necessary protection (Manandhar et al., 2018). Good ventilation with low humidity and propertemperatures must be maintained at the storage sites of grains (Uniyal, 2014). Generally, storage temperature below 41^{o}C is recommended to minimize bacteria, fungal, and mold growth, whereas a temperature above 70^{o}C increases their outbreak(Arahet al., 2016).

Table 1 Storage compatibility of agricultural crops

Group 1. Temperature 32-36 ºF, Relative Humidity 90-95%				
Apples*	Berries	Grapes	Pears*	Rutabagas
Asian pears*	Cantaloupe*	Parsnips	Plums*	Turnips
Beets, topped	Cherries	Peaches*	Radishes	
*****These items can produce high levels of ethylene that can be detrimental to items in Group 2.				
Group 2. Temperature 32-36 ºF, Relative Humidity 90-95%				
Beets, topped	Cabbage	Cherries	Greens	Radishes
Berries	Carrots	Corn	Lettuce	Rhubarb
Broccoli	Cauliflower	Grapes	Parsnips	Rutabagas
Brussels sprouts	Celery	Onions, Green	Peas	Turnips
Group 3. Temperature 32-36 ºF, Relative Humidity 65-75%				
Garlic	Onions	Shallots		
Group 4. Temperature 50 ºF, Relative Humidity 90-95%				
Beans†	Eggplant	Peppers	Squash, Summer	Tomatoes, Ripe
Cucumbers	Okra	Potatoes†	Squash, Winter	Watermelon
†Fifty degrees is slightly above ideal conditions for these commodities.				

Source: Boyhanet al. (2009).

3.1.5. Processing

Processing includes postharvest activities such as freezing, drying, milling, canning, mixing/fortifying, fermenting, sprouting, dehulling, and adding salt, sugar, fat, or additives after harvest (Greenberg, 2017). According to Greenberg (2017), it is used to produce food that is safe, diverse, abundant, and accessible in supply. Mainly, processing methods such as fortification and fermentation add nutritional value. However, consuming an excessive amount of highly processed food results in lower-quality food (Mozaffarian et al., 2011). During processing and packaging, intentional losses, inefficiencies in techniques and malfunctioning, deviations during processing lines, restriction because of legislation, extra production and the packaging system should be minimized (Ishangulyyev et al., 2019).

Biofortification also increases the concentration of specific microelements, lowers the concentration of phytate, or increases the activity of compounds that improve the digestibility of essential elements and compounds by adding

zinc, iodine vitamins, and iron either at the time of processing or processing at the household level (Kruger et al., 2020). Amino acids in biofortified plant foods, for example, improved zinc digestibility or availability (Gharibzahedi et al., 2017). A molecule Nicotianamine is a metal chelator in transgenic plants manipulated by biofortification, which improves intestinal Fe and Zn absorption by enhancing the filling of Fe and Zn into certain foods such as grains (Borrill et al., 2014). It also improves conventional fortification because it provides nutrients to the rural poor who do not have access to fortified foods (jBouis and Saltzman, 2017). The use of biofortified staples reduced the prevalence and duration of diarrhea in children aged above 5 and 3 years by 11.4% and 18.9%, respectively (Jones and de Brauw, 2015). Some countries use vitamin and microelement fortification of foods. Some of the biofortified crops and foods are indicated in Table 2.

Table 2. Some biofortified crops with their improved characteristics

Biofortified crop	Biofortified by	Reference
Orange fleshed sweet potato	Vitamin A	De Brauwet al. (2018)
Maize and wheat	Zn and Se	De Valençaet al. (2017)
Bean and pearl millet	Iron	Haaset al. (2017)
Pearl millet flatbread	Iron	Finkelsteinet al. (2015)
Provitamin A maize	Vitamin A	Palmeret al. (2016)
Yellow cassava	Vitamin A	Talsmaet al. (2016)
Wheat	Zink	Kinget al.(2015)
Biscuits	Iron	Parasar and Bhavani (2018)

Fermentation is the mechanism by which food loss and waste are minimized. It also helps in reducing the amount of time and fuel required for cooking, which increases food production and sales (Waqas et al., 2018). During fermentation, the development of lactic acid bacteria enhances intestinal tract health, the immune system, synthesis, and bioavailability of nutrients; decreases lactose intolerance, susceptibility to allergy, and cancer risks (Parvez et al., 2006). Furthermore, fermentation increases prebiotic availability, extends shelf life, prevents pathogenic contamination, reduces anti-nutrients (Knockaert et al., 2014), reduces lactose, and increases lactase enzyme for improved digestibility in dairy products (Savaiano, 2014).

Germination is the other ordinary bioprocessing technique used to develop modified grain and legume flours with improved nutrient qualities and solubility (Xu et al., 2020). Malting amaranth, for example, increased in-vitro protein digestibility, protein composition, oxalate and tannin content while decreasing phytic content (Hejazi et al., 2016). Similarly, phenolic bioactive compounds were increased during quinoa sprouting, with antioxidant capacity, phenolics, and flavonoids increasing by 40, 49, and 62%, respectively. Phenolic bioactive compounds found in sprouted cereals can control chronic hyperglycemia and oxidative stresses in people with type 2 diabetics. Sprouting also improved the functionality of wheat-based bread flour (Aguilar et al., 2019).

Pickling has been used to preserve fruits, vegetables, fish, and meat. Moreover, probiotics improve the safety of preserved foods and reduce pH through produced acids and detoxification during pickling (Behera et al., 2020). Drying is also used to remove the water content of foods through the use of evaporation, yielding solid products for long-term storage. For instance, drying to less than 10% water content inhibits microbial growth, and is used to extend shelf life. It is also used to reduce the water content and increase the concentration of the components contained in food products. It also saves storage space and shipping costs (Djaeni and Perdanianti, 2019).

3.1.6. Packaging and packing of processed products

Food packaging helps to prevent the products and ensure a good distribution of food products without physical damage, chemical contamination, or biological and environmental hazards that can threaten life (Wyrwa and Barska, 2017). It also plays a major role in improving nutrition by extending shelf life and facilitating the wider distribution of processed foods, during packaging and packing, caring is necessary to reduce loss at storage sites (Arah et al., 2016). Therefore, packaging material should fulfill the fundamental roles of packaging, such as protecting/preserving food, providing convenience, and informing the consumer to reduce food waste and protect the consumer's health (Wyrwa and Barska,2017).

3.1.7 Food labeling

CODEX(1991)defined food labelling as any written, printed, or graphic information expressing the food or presented near the food together to promote the sale of the food or why the food is disposed of. Words, letters, logos, images,

and symbols refer to the shelf life, method of preparation, consumption, nutrient quality, and commercial characteristics of a product (Wyrwa and Barska, 2017). Any information on the label should be accurate, standardized, appealing, and simple for consumers of all literacy levels to understand (Darkwa, 2014). Furthermore, the supervisory authority must verify the label in order to reduce declarations that mislead consumers (Wyrwa and Barska, 2017).

3.1.8.Retail distribution

The retail market is the ultimate destination for agricultural products and visible crop handling practices.Consumers can choose to buy or reject food in packaging materials or unpackaged products. The design of the commodity display should allow for quick and impulsive purchases (de la Pena et al., 2018). The quality of commodities at the retail distribution stage is determined by factors such as incompatible commodities displayed close together, period of exposure, temperature, relative humidity, lighting, and how to store personnel and consumers handle the produce (Kumarand Kalita, 2017). Inadequate conveyance conditions, poor transportation and market facilities, insufficient roads, business rules, consumer perceptions, and packaging preferences are also causes of loss and waste during retail distribution. Therefore, rapid turnover is recommended as a food and loss reduction mechanism at this stage (Ishangulyyev et al., 2019).

3.1.9. Safety and quality of food products

Food safety is the assurance that food will not cause harm to the consumer when prepared and consumed as intended.Biological contaminants such as bacteria, viruses, parasites, and prions, as well as chemical hazards such as heavy metals, environmental pollutants, mycotoxins, and allergens, can contaminate foods (WHO, 2019). According to FAO (2019), food-borne spoilage causes six million illnesses, four hundred and twenty million deaths, and thirty-three million disabilities. The food-borne disease affects 40% of children under the age of five. Detection and abolishing the risks of food borne illness are difficult because of interconnected global markets, the length of supply chains, and the growing number of stakeholders in the food system. Food quality, hygiene, and safety standards are required in such cases to protect consumers and improve access to safe and nutritious foods (Khairuzzaman et al., 2014).

3.2. Appropriate postharvest handling practices of durable crops

Several factors, including biological and environmental factors, limitations in mechanization accessibility and cost, a poor marketing system and farmer organizations, lack of information, insufficient irrigation water and infrastructure, a lack of credit, inadequate improved varieties, and an inadequate policy framework and governmental regulation, all pose challenges to nutrition-sensitive postharvest handling practices (Bolarin and Bosa, 2015). Postharvest losses in cereal grains were primarily caused by inefficient harvesting, drying, threshing, winnowing, storage, transportation, packaging, and marketing (Mutungi et al., 2020). In Uganda, for example, estimated maize postharvest losses accounted for 6.4 16.4%, 4.0%, 1.3%, 2.4%, and 1.2-5.9% at the harvesting, drying, shelling, transport to farm, and storage stages, respectively. Gender disparity in the value chain is another factor of postharvest crop loss, and findings also confirmed that female-headed farmers experienced lower crop loss (Shee et al., 2019). Similarly, the degree of loss is determined by the level of education and training received by the farmers. In Ethiopia, Tanzania, and Uganda, for example, postharvest losses are reported to be lower for more educated and trained farmers than for less educated and untrained farmers (Shee et al., 2019). Generally, appropriate postharvest handling reduces loss, increases producer income, lowers consumer prices, increases food availability, creates job opportunities, and improves the livelihoods, food and nutrition security of rural and urban consumers (Mezgebe et al., 2016). The following topics explain some of the appropriate handling practices at each postharvest value chain stage.

3.2.1. Pre-harvest drying

The cereals or legumes should be dried during pre-harvest drying to prevent moisture content. Furthermore, avoiding prolonged pre-harvest field drying may increase the risk of loss due to birds, rodents, insects, moulds, and theft. The crop should also be harvested at the proper maturity stage to reduce seed decay due to mould development (Njoroge et al., 2019).

3.2.2. Harvesting and transporting

Harvesting is the point at which the grain's maximum quality should be achieved. Harvesting time is determined by the degree of maturity of the grain. However, the lack of a proven maturity index for some crops and native export markets, lower adoption of established indices due to price and distance to market, poor weather conditions at harvesting time, and moisture content of harvested products cause grain loss during harvesting (Bolarinand Bosa,

2015). A large number of losses occur before or during harvesting operations if they are not performed at optimum crop maturity and moisture content because too early crop harvesting increases crop drying costs and grain susceptibility to mould, insect, and breakage during milling (Emana et al., 2017). Shattering, birds, rodents, animals, thefts, rain, and storms, on the other hand, cause losses if the crop is left on the pitch past its optimal maturity (Kuyuand Tola, 2018). Furthermore, a lack of sufficient labourers during peak harvesting delays harvesting, resulting in significant losses (Coker and Ninalowo, 2016). Therefore, an appropriate harvesting stage, method, and time, as well as well-designed harvesting tools, equipment, and harvesting containers, are suitable for reducing qualitative and quantitative loss during subsequent postharvest handling practices (AFC, 2018). It is also necessary to prevent grain from falling on the road when transporting harvested crops from field to threshing or storage location (Kumarand Kalita, 2017).

3.2.4. Postharvest drying

The time required for complete drying is determined by atmospheric and weather conditions. Unprotected drying and threshing places make grains vulnerable to livestock, birds, rodents, and small ruminants at this stage. Contamination of the grains by the droppings of these animals results in higher losses and is a nutrition-sensitive postharvest handling problem (Asemu et al., 2020). Brittleness and cracking of grains after threshing, hulling, and milling also result in significant postharvest losses. Consequently, the broken grain will be separated from the husks, resulting in weight loss and a decrease in the price paid for the grain when it is sold. Therefore, during postharvest drying, the surface should be covered or raised in drying areas with accurate techniques for assessing the grain moisture content (Shee et al., 2019).

3.2.5. Threshing

Before threshing, the crop should be sufficiently dried because threshing will be futile if the crop is not completely dried. Therefore, proper management of grain moisture content is vital before threshing. Lack of proper threshing equipment isalso the causes of losses in grains (Coker and Ninalowo, 2016). Grains should also be thoroughly cleaned to prevent insect infestation and mould growth during storage. Furthermore, proper grain cleaning during threshing helps to reduce the development of unwanted taste and colour as well as damage to processing equipment during processing. The area for threshing or grain storage should also be large enough to hold the grains and be clean (Kumarand Kalita,2017).

3.2.6. Storage

In developing countries, cereal storage accounts for the majority of postharvest losses (Kumar and Kalita, 2017). Traditional storage structures are used to store grains in these countries. Traditional storage structures, however, are incapable of completely preventing insect infestation and mould growth in storage, resulting in significant losses. Ethiopian farmers, for example, mechanically remove insects, infested grains or cobs, shake, and restack the grains to disturb and reduce the activity of the insects. Farmers also heat the grains by exposing them to the sun, treating them with cow urine, and admixing salt during storage to control sorghum infestationduring storage (Mendesil et al., 2007).

Appropriate, low-cost and simple loss reduction mechanisms are reported for low-income countries. For instance, Mohapatra et al. (2015)suggested using low-cost, low-risk, locally available materials such as inert dust and hermetic storage materials to control insects in storage. Proper use of these mechanisms prevents insect infestation, ensures effective resource utilization, increases production and use, family income and standards, and improves food and nutrition security in households (Kembabazi, 2019). When compared to traditional storage materials, hermetic storage such as metal silos and grain bags reduces aflatoxin B1 contamination, kills insect adults, and reduces insect infestation during maize storage. For example, Kumar and Kalita (2017) reported that proper hermetic storage materials can reduce infestation by up to 98%.

In general, adequate storage facilities, personnel hygiene, and effective and sufficient monitoring are required during grain storage to control an infestation of insects and mould, rodent and bird entry, temperature, moisture, and humidity (Kumarand Kalita, 2017). For example, storing grain at a temperature of less than 15^0C and a moisture content of 9% to control mould and insect infestation in grain storage is highly recommended (Befikadu, 2014). Moreover, good agricultural practices and appropriate storage technologies can significantly reduce losses and contribute to food security, increased farmer income, and poverty alleviation (Kumar and Kalita, 2017). Using recommended storage pesticides at their recommended levels is also beneficial in limiting insect and mould infestation of grains during storage.

3.2.7. Food losses and wastes prevention

Food waste refers to food materials and beverages that are intended to feed humans but were not ultimately consumed by humans(Gustavsson et al., 2011). It also refers to food that suffers unintentional deterioration in quality or quantity as a result of food spills, spoils, bruising, wilting, or other damage caused by infrastructure limitations (Papargyropoulou et al., 2014). It reduces food volume, quality, and nutrient losses, resulting in lower nutritional content (Timmermans et al. 2014). It also raises the cost of unnecessary transportation, energy, water, fertiliser, machinery and equipment, packaging, labour, and capital (FAO, 2014). Therefore, interventions that reduce food loss and waste must be implemented throughout the food chain.

4. Women empowerment in Nutrition-sensitive agriculture

Women are the distinct central point in nutrition and agriculture because they play economic, production, processing, marketing, vending, selecting, consumption, and preparation roles in their households (de la Pena et al., 2018). Therefore, empowering women is critical for proper time allocation for child feeding and care, farming, and working in the home to improve food and nutrition security for their families. Furthermore, giving women equal access to productive resources as men, the production yield increased by 20-30%, and a decline in 100-150million was recorded for malnourished (Quisumbing et al., 2014). Women control income and resources in families and have market access; therefore, they influence household food, nutrition security, children's dietary intake, and child education. Furthermore, involving women in income-generating activities improved their economic, social, and nutritional status while decreasing gender inequality (Shafiq et al., 2019). Likewise, child stunting is lower in families where women are empowered and educated (Jones et al., 2019). Overburdening women in food production and postharvest activities, on the other hand, may affect the time they devote to caring for their babies and food preparation (McNamara and Wood, 2019; Shafiq et al., 2019). Therefore, careful consideration of their allocation of time and management over resources, societal rank, and responsibilities in and outside their home is vital to confirm that developing nutrition-sensitive value chains that encourage women's empowerment is beneficial to achieve positive nutrition outcomes (de la Peña et al., 2018).

5. The overall potentials and missing links in food and nutrition security

Achieving the Sustainable Development Goals (SDGs),mainlyGoal number two in developing countries by 2030, is highly challenging. Thus, the efforts of researchers and policymakers in identifying many potentially valuabletechnologies are paramount. Various postharvest loss and waste reduction mechanisms are being developed, and in some cases, public awareness is being raised (Ishangulyyev and Lee, 2019). Good agricultural and manufacturing practices are well-identified and understood measures. Likewise, the contribution of underutilized food types (Aderibigbe et al., 2020; Minde et al., 2021), indigenous local foods (Kimani et al., 2020), agro-forestry (Jemal et al., 2018), and mixed and integrated crop production (Gideon et al., 2021) was confirmed in food and nutrition security. However, stakeholders in the food system must be aware of the factors that can impact food and nutrition security. The stakeholders must also discuss climate change's impact on food and nutrition security (Durst and Bayasgalanbat, 2014). Despite the fact that researchers and policymakers identify potential technologies for food and nutrition security, the cost and benefit of the technology are not well known. Besides, because there are no close relationships between laboratory work reports and its extension, the technologies developed are difficult to apply in most low-income countries because the researchers do not consider the local cost. When farmers are given training, stakeholders do not check and assist them when they want to apply the information (Uccello et al., 2017). Even the mechanisms for combating them are lacking, and the expected results are inadequate due to a lack of adequate market access, transportation, necessary tools, awareness, governmental regulations, and legislation (Beddington et al., 2012).

Though agroforestry provides vital benefits in food and nutritional security, increases agricultural productivity, mitigates the effects of climate change, and increases environmental flexibility, and failure to extend advanced agricultural methods hinders smallholder farmers in developing countries from reaping the full benefits of agroforestry (Jemal et al., 2018). Furthermore, the lack of legislation and certification makes it difficult to promote and use underutilized indigenous foods and technologies (Rampa et al., 2020). The attention given to wild fruits and vegetables is also not as good as that of agricultural products. In areas where water is scarce, solar-powered irrigation technologies are not practiced.Researchers and policymakers ignored changes in the level of indigenous knowledge and contribution of human labor, which are critical in food security and require the same attention as agricultural sectors (Kuiper et al., 2020). In the areas where some politicians can use food as a weapon, people can starve as violations lead to food insecurity and food insecurity, which worsens the conflict (FAO/WFP, 2022).

Conclusion

Nutrition-sensitive agriculture and appropriate crop postharvest handling are widely recommended interventions in addressing food security, particularly in low-income countries. The management system used during production affects crop quality and/ or safety. The postharvest actions conducted on the crop also influence its quality and/or safety. Therefore, if implemented correctly for agricultural products throughout the value chain, these two interventions have the potential to control all types of malnutrition. Nonetheless, these interventions are hampered by a lack of complete agricultural input packages, facilities, awareness, and income. Therefore, coordinated and effective management of handling, storage, transportation, and methods and tools appropriate to gain maximum benefits from agro-forestry in various human and ecological settings is critical. To get the most out of agriculture, it is also important to raise awareness about nutrition outcomes, empower women, and promote mixed farming systems.

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The study comparison was between the Small East African Zebu and the Tyrolean Grey cattle crosses in Uganda

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Abstract
This study was conducted to assess farmers' perception of the performance of indigenous Small East Africa Zebu (SEAZ) and Tyrolean Grey (TG) x SEAZ cattle cross. TGZ cattle is indigenous to the alpine region of Austria. Cross-sectional survey was carried out among 45 respondents comprising 37 farmers, three (03) artificial insemination technicians, three (03) extension workers, and two (02) farm managers in the Ugandan districts of Kole, Nakapiripirit and Kayunga. Source of labour ($\chi2=6.042$, $p<0.05$) influenced farmers' preference of cattle. Traits such as better draft power ($\chi2=6.943$, $p<0.01$), resistance to parasites and diseases ($\chi2=4.477$, $p<0.05$) were significantly associated with SEAZ. On the other hand, docility ($\chi2=4.847$, $p<0.05$), better milker ($\chi2=13.976$, $p<0.001$), good mothering ability ($\chi2=10.174$, $p<0.001$), fast growth ($\chi2=11.242$, $p<0.001$) and hard hooves to withstand muddy places better ($\chi2=4.498$, $p<0.05$), were significantly associated with TGZ. More than three quarters (78.8%) of the farmers preferred the TGZ because they felt that it performed better than the SEAZ
Keywords: Productivity, traits, Perception, preference, significance, semen

1. Introduction

Agriculture is a critical sector of Uganda's economy, contributing about 24.6 percent to GDP and 71 percent to employment (World Bank, 2019). Crop production and livestock farming are important (MAAIF, 2016). Uganda is a low-income country with a population of 40 million (UN, 2018) and a GDP per capita of USD 607 per year. As a way of increasing agricultural productivity, the government of Uganda ensured continued investment in technology improvement through research for improved breeds (Uganda Vision, 2040). The Small East African Zebu cattle (SEAZ) is a dual purpose breed found mainly in the Eastern and Northern parts of Uganda. Although it is well adapted to harsh environmental conditions (Wurzinger *et al*, 2014) and the prevalent low-input low-output production system, its poor performance in terms of low meat and milk production, late maturity and small size are becoming a concern to farmers who would like animals with higher productivity.

In 2009, the Tyrolean Grey Cattle Breeders' Association (TGBA) donated 1000 doses of Tyrolean Grey (TG) cattle semen to the National Animal Genetic Resources Centre and Databank (NAGRC&DB). NAGRC&DB is a government agency mandated to spearhead livestock breeding in the country. More donations of 1000 TG semen straws to The National Livestock Resources Research Institute (NaLIRRI), an institution under the National Agriculture Research Organization (NARO), and NAGRC&DB by TGBA followed in 2015. TG semen was first used on the SEAZ cattle that are kept at Lusenke Stock Farm,

Kayunga District (Central Uganda) and among farmers in the neighborhood of the farm. This farm, owned by NAGRC&DB, is at the forefront of conserving SEAZ and one of its conservation strategies is to cross the SEAZ with better performing breeds.

The TG cattle is a dual purpose breed that originates from Austria and is used in typical mountain farming under rough conditions. They are among the few small bodied temperate breeds that could comfortably be used to cross with the small bodied SEAZ with no threat of dystocia. The SEAZ has over the years survived the introduction of exotic genetics that would have enabled it to achieve rapid progress in milk yields and daily growth performance like other temperate breeds. However, the lag in performance is highly compensated by its high adaptability to extreme environmental conditions. The study objectives was to assess the socio economic characteristics of the cattle farmers, influence of socio- economic characteristics on breed type preferences and determine farmers perception of the performance of SEAZ and TGZ.

2. Methods

Description of the study area

The study was carried out in five districts of Uganda (Fig. 1) where the TGZ are reared. The first location, was Akuyam parish in Nakapiripirit District. The coordinates are Longitude 34.7216^{o} E Latitude 1.8503^{o} N. It lies at an altitude of between 1,356-1,524m above sea level in a semi-arid area with temperature average of 30^{0}C per annum. The second study area was in Abeli and Agege in Kole district in northern Uganda. The study areas have the following coordinates: Abeli at Longitude 32.6307E 32^{o}38’3.458”, Latitude 2.19564 N 2^{o} 11'44.6316. It lies at an altitude 1061.8m above sea level. Agege is at Longitude 32.7839 E 32^{o}47”1.986”, Latitude 2.21902 N 2^{o} 13’8448. It lies at an altitude of 1056.58m above sea level. Third study area was Kayunga district located in central Uganda, and the study there was conducted at Lusenke stock farm. The coordinates are Latitude: 0.9822687 and Longitude: 32.9302079. Kayunga practices mainly 2 types of agriculture that is animal husbandry and crop husbandry. The fourth study area was Buikwe district in central Uganda. The study was conducted at Njeru stock stock farm with coordinates as Latitude: 0.4196434 and Longitude: 33.185576. The fifth study area was in Wakiso district located in central Uganda. The study was conducted at Gayaza High School Farm, with the coordinates as Longitude 32.7839 E 32^{o}47”1.986 and Latitude 2.21902 N 2^{o} 13’8448.

Figure 1: Map of Uganda showing the Study area

Sampling and Data Collection

This study adopted a cross sectional survey research design. It used mixed approaches that included both qualitative and quantitative methods to determine the influence of demographic factors on farmers' perception of the preferred traits and breed. Purposive sampling technique was used to identify and recruit all the 37 farmers rearing SEAZ and TGZ cattle (Fig. 2), three (03) artificial insemination (Al) Technicians, three (03) extension workers, and two (02) farm managers, totaling to 45 respondents. Primary data were collected from respondents by use of pre-tested semi-structured questionnaire, individual in-depth interviews, focus group discussion, and personal observation.

To assess the preferred traits and breed, performance indicator traits included: ability to walk for long hours, resistance to parasites and diseases, good draft power, survival during drought, good milker, docility, good mothering ability, high calf survival, fast growth rate, and hard hooves to withstand muddy places. The aesthetic traits included coat colour and horn shape. In order to understand the context in which the study was done, elements of the socio-economic characteristics of the respondents were also captured.

Figure 2: Tyrolean Grey cattle crosses (TGZ) and Small East African Zebu (SEAZ)

Data Analysis

Data collected was subjected to field and desk editing to ensure accuracy, consistency and completeness. The data was then entered into Microsoft Excel and exported to SPSS (version 20) and STATA (version 15) for cleaning and eventual analysis. Descriptive statistics involved the use of central tendency (mean), proportions, and standard deviations. The inferential tests employed the use of chi square test, to test the association between the main study variables; as well as to test the hypotheses. These were applicable to the association between the socio-economic characteristics and preference parameters. All tests were measured at 95% confidence interval.

3. Results and Discussion

Socio-economic characteristics of the cattle farmers

The key findings from socio-economic characteristics (Table 1) revealed that, more than three quarters of the respondents (95.6%) were male while 4.4% were female and this show that; most of the farm managers and/or farmers rearing the SEAZ x TGZ were mainly male. This implied that, there was gender imbalance as observed from the percentage of men involved in the study compared to women. Men always dominate and women do most of the work in terms of rearing, milking and treatments for the animals that fall sick yet goals and priorities of men and women in the households differ (MAAIF, 2016).

Table 1: Socio-economic characteristics of the cattle farmers

	F	Mean (±SD)
Gender of respondent		1.04 (0.208)
Male	43	
Female	2	
Age group of respondents		42.68 (13.761)
35 years and below	15	
36-50	15	
Above 50 years	11	
Education level of respondents		2.89 (1.603)
No formal education	11	
Primary	12	
Secondary	5	
Tertiary	3	
University	13	
Marital status		1.10 (0.379)
Married	37	
Single	2	
Widowed/separated	1	
Purpose of owning land*		1.21 (0.415)
Crop production	26	
Grazing and forage production	25	
Labour source for animal keeping		1.65 (0.834)
Family labour	23	
Hired labour	8	
Both family and hired	9	

Values in parenthesis are percentages and standard deviations, SD=Standard deviation. Multiple response analysis was allowed, case by case analysis and missing value excluded.

The results from (Table 1) revealed that; slightly more than a quarter of the respondents (26.8%) were over 50 years of age. Equal proportions of the respondents (36.6%) were between 36-50 years of age and below 35 years of age, respectively. This finding implies that most of the respondents rearing the SEAZ and TGZ were of middle age and youth. The study area had relatively better potential of economically active population who could participate in dairy cattle production. These results agree with those of Kafle and Shah (2012) who observed a positive and significant relationship between age and technology adoption.

Table 1 indicate that, more than a quarter of the respondents (29.5%) had university education and training on dairy production, another (27.3%) had attained primary education level and a quarter of the respondents (25%) had no formal education, while (11%) had attained secondary level education. This implied that the farmers rearing the TGZ had some level of literacy and have some reasonable knowledge of taking care of the cattle breeds. The average years spent by the majority of cattle farmers in school were relatively high. This means that education plays an important role in livestock production. With more years spent in school, they are better positioned to recognize the importance of crossbred animals.

Like the current result, according to Lemma (2017), education levels of household heads have impacts on potential of milk production. The more educated a household head is, the more he/she is likely to acquire, understand, obtain, disseminate new technologies within a shorter time and more efficient when allocating resources compared to a household head with fewer years of education (Kafle and Shah 2012; Ebojei *et al*. 2012).

A large proportion of the respondents (85.7%) were married. This could be the reason why more males participated in this study because they are the decision makers. The results from (Table 1) revealed that, land owned was majorly for purposes of crop production (51%). However, some (49%) of the respondents also owned land for the purposes of grazing and forage production The results revealed that, the major source of labour for keeping the animals was family labour (57.5%), however, close to a quarter of the respondents (22.5%) used both family and hired labour while, some (20%) of the respondents used only hired labour (Table 2). Family labour constituted the highest percentage. The reason could have been to reduce the cost of labour payment and proper farm management. This was an indication that dairy cattle management requires the attention of family members since they have high value. In line with the current results, Megersa et al. (2011), and Gillah (2012) reported the same result from different parts of Ethiopia.

Influence of socio-economic characteristics on breed type preferences

The independent sample t-test and chi square (Table 2) were used for continuous and categorical socio-economic variables respectively. All the socio -economic variables did not significantly influence farmers' preference as regards the performance of the SEAZ or TGZ except Source of labour (χ^2=6.042, p<0.05). This could have been because dairying is labour intensive. According to (Camara *et al.* (2019), without a good understanding of these factors, it would be very difficult to persuade the local farmers to fully participate and cooperate in a breeding programs.

Table 2: Influence of socio- economic variables on farmers' breed type preference

Variable (n=45)	Mean (±SD)		T (p-value)	Chi square (p value)
	SEAZ	TGZ		
Gender of respondent	1.00 (0.000)	1.06 (0.236)		0.598 (0.439)
Age of respondent	44.4 (12.886)	42.13 (14.191)	0.472 (0.643)	
Education level of respondents	1.90 (1.287)	3.18 (1.585)		5.847 (0.211)
Occupation of the respondent	1.30 (0.483)	2.28 (1.689)		3.287 (0.511)
Marital status	1.00 (0.000)	1.13 (0.434)		1.081 (0.582)
Purpose of owning land	1.00 (0.000)	1.29 (0.464)		3.332 (0.068)
Source of labour for rearing animal	2.00 (1.414)	2.46 (0.776)		6.042 (0.049)*
Household size	8.20 (4.077)	6.36 (3.332)	1.248 (0.232)	
Own land	1.00 (0.000)	1.16 (0.374)		1.837 (0.175)
Hectares owned	8.30 (6.533)	15.04 (38.443)	0.862 (0.396)	
Years of keeping animal	6.89 (8.936)	3.92 (2.261)	2.263 (0.057)	

**** p<0.001, ** p<0.01, *p<0.05*

Preferred traits of the SEAZ x TGZ

The findings from (Figure 3) revealed that, the SEAZ scored (68.3%) for ability to walk long hours as compared to the TGZ (38.6%). These breeds have unique genetic attributes such as adaptation and tolerance to drought, heat, diseases and ability to utilize low-quality indigenous forages. According to Nalule, (2010) pastoral communities select breeding cattle with attributes to withstand challenges created by landscapes. This is common in semi-arid region of Karamoja where cattle have to trek long distances in search of pastures and water. The results from (Figure 3) revealed that, the SEAZ scored (85.4%) for resistance to parasites and diseases as compared to the TGZ (40.9%).

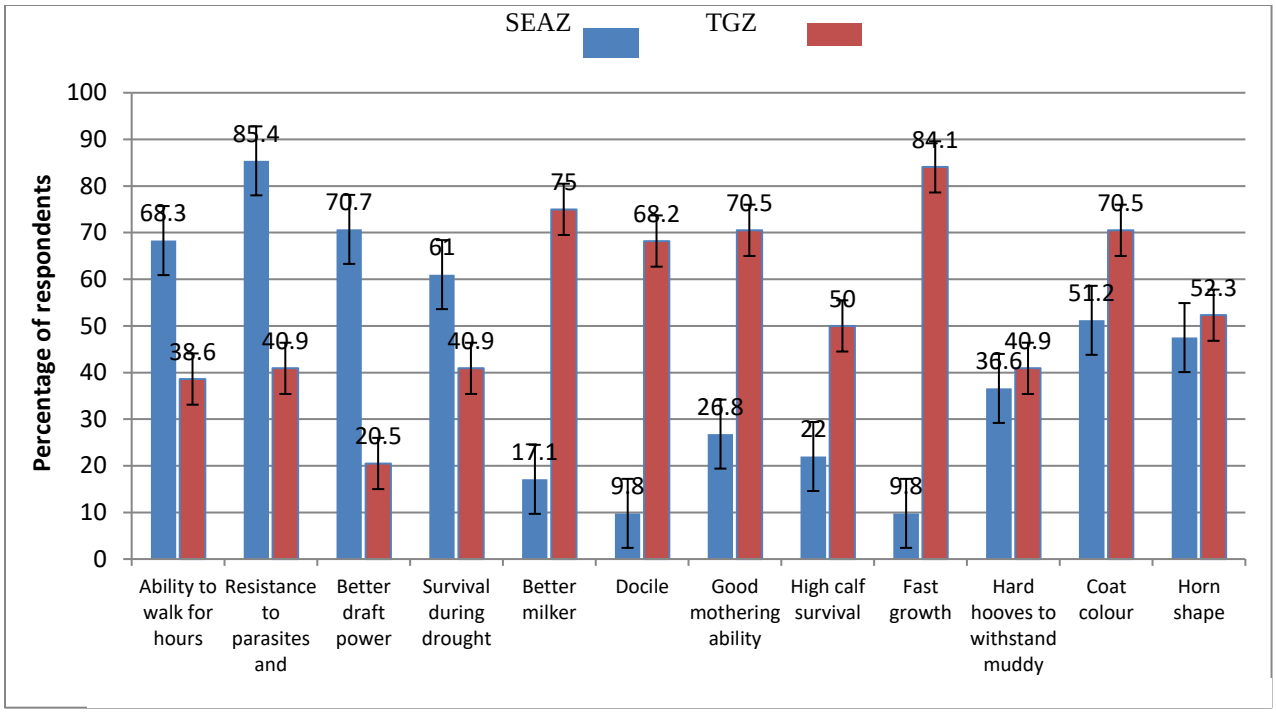

Figure 3: Preferred traits of the SEAZ x TGZ

Percentages of farmers that preferred a certain breed (SEAZ or TGZ) based on preferred traits are presented;

This result agrees with that of Hiemstra (2015) who observed that many farmers who keep dual-purpose breed are pleased with the good resistance of their herd and the low treatment costs. Traditional management practices, different landscapes, Socio-cultural needs and endemic disease challenges have over time enhanced indigenous cattle breed adaptation to their localities. The adaptations to diverse Agro-Ecological Zones take the form of variations in body sizes and levels of productivity (Kugonza et al., 2011), morphometric traits variations (Kabi et al., 2015) and resilience to endemic diseases (Magona, Walubengo and Kabi, 2011; Kabi et al., 2014). According to Bishop (2012), several studies have indicated that indigenous cattle can endure and be sustainably productive in the presence of disease challenges, a phenomenon referred to as tolerance or resilience to disease and endemic stability to infection. Jonsson et al., (2012), have also noted that continuous exposure of endemic diseases such as tick-borne diseases among indigenous cattle populations has led to the development of tick-borne disease tolerant traits.

Most respondents reported that, the SEAZ scored (70.7%) for better draft power as compared to the TGZ (20.5%). According to Murage and Ilatsia *et al*. (2011) reasons for rearing animals are; animal traction, reproduction, symbol of wealth, security, dowry payment, employment, prestige, and as a shield against inflation. The results in (Figure 3) showed that, the SEAZ scored (61%), for survival during drought as compared to the TGZ (40.9%). This is in line with the findings of Jaritz (2014) who indicated that animal breeds were originally selected according to their functions, and only those that adapted best to the environment, husbandry conditions, and the demands of their holders survived.

The study in (Figure 3) showed that, the SEAZ scored (61%), for survival during drought as compared to the TGZ (40.9%). This is in line with the findings of Jaritz (2014) who indicated that animal breeds were originally selected according to their functions, and only those that adapted best to the environment, husbandry conditions, and the demands of their holders survived. For example, in the semi-arid region of

north eastern Uganda, the climatic conditions vary from arid to semi-arid with seasonal availability of pasture and water (Nalule, 2010), to which traditional Karamojong shorthorn zebu cattle are well adapted.

As revealed in (Figure 3), the TGZ scored (75%), for better milker as compared to the SEAZ (17.1%). the SEAZ (17.1%). Milk production was the most important trait that farmers considered when selecting breeds. This is probably because milk production for feeding the family and for generating cash income were the reasons for rearing the TGZ. According to Usman *et al*., (2013), in addition, consumers' increasing demand for superior quality food and regional products indicates that there are open markets for such products (Warschum et al.; 2013). Whereas much of the expansion of livestock production in Africa have been in form of increased stocks of animals (FAO, 2015), strategies to transition to more productive livestock production systems have been shown to be helpful in simultaneously reaching desirable outcomes on climate and food availability (Havlik et al., 2014). The results from (Figure 3) revealed that, the TGZ scored (84.1%) for fast growth as compared to SEAZ (9.8%). Fast growth rate can be of advantage to farmers aiming at producing heifers for sale. The preference for high growth rate of calves might be related to early breeding of heifers, because the farmers are still in the process of building their dairy herds. Besides getting a better price for old dairy cows, farmers receive also a higher price for the bull calves when sold to a fattener because they grow faster (Geuder et al., 2012).

The results from (Figure 3) revealed that, the TGZ scored (68.2%) for docility as compared to the SEAZ (9.8%). In the pastoral production system, farmers termed "TGZ as mental" because they are so docile that, they keep following herds'men wherever they go, while in the semi-intensive production system, it was reported that they are aggressive. This could be because of rearing them with large animals like the Friesians. Docility therefore as a dairy cow trait, allows for ease in handling and, by extension, ease in milking (Gergovska et al., 2014). The results from (Figure 3) revealed that the TGZ scored (70.5%) for good mothering ability as compared to the SEAZ (26.8%) The dual-purpose breeds are not selected exclusively for milk production; dual-purpose cows have a more diverse genotype. In general they show better health, higher fertility and a better longevity than the Holstein Friesian (Piccand et al., 2013).

The TGZ scored (70.5%) for coat color and (52.3%) for horn shape as compared to the SEAZ at 51.2% and 47.5%, respectively. This result agrees with that of Nalule (2010) who observed that specific features of the SEAZ such as size and shape of horns, multiple colourations, and body size are carefully selected for by kraal leader through utilization of specific sires and dams. Horn shaping and spacing are also critical traits in enabling cattle to graze in thickets and difficult terrain (Kugonza et al., 2012b). The results from (Figure 3) revealed that the TGZ scored (40.9%) for hard hooves to withstand muddy places better as compared to the SEAZ (36%). However, the TGZ was cited with foot rot on one of the farms visited due to floods in the pasture land. The results from (Figure 3) revealed that, the TGZ scored (50%) for high calf survival as compared to the SEAZ (22%).

Relationship between preferred cattle breed type based on preferred traits

The chi- square test showed that, trait such as; better draft power (χ^2=6.943, p<0.01) was significantly associated with SEAZ. This could have been because the TGZ are new breed that have been introduced in Uganda and most farmers have not tried to use them for ploughing. This indicated that livestock development interventions should be geared towards breeding of animals that will produce draft power. Other authors have reported similar findings, that the use of cattle for draught purposes was a major activity in the agro pastoral systems (Traoré *et al*., 2016).

Table 3: Testing for the relationship between preferred cattle breed type and preferred traits

Preferred traits	χ^2	Df	Sig
Ability to walk for hours	2.354	1	0.125
Resistance to parasites and diseases(SEAZ)	4.477*	1	0.035
Better draft power (SEAZ)	6.943**	1	0.008
Survival during drought	1.547	1	0.214
Better milker(TGZ)	13.976***	1	0.000
Docile(TGZ)	4.847*	1	0.028
Good mothering ability(TGZ)	10.174***	1	0.001
High calf survival	0.335	1	0.563
Fast growth (TGZ)	11.242***	1	0.001
Hard hooves to withstand muddy places better(TGZ)	4.498*	1	0.034
Coat colour	1.964	1	0.161
Horn shape	3.333	1	0.068

**** p<0.001, ** p<0.01, *p<0.05*

Multiple responses were accepted, case by case analysis and missing value excluded

Resistance to parasites and diseases (χ^2=4.477, p<0.05) was significantly associated with the SEAZ. Therefore, an animal showing resistance to ticks and diseases becomes more profitable to the low-input production system. This result agrees with that of Hiemstra, (2015) who observed that many farmers who keep dual-purpose breed are pleased with the good resistance of their herd and the low treatment costs.

Milk production was the most important trait that farmers considered when selecting breeds. Better milker (χ^2=13.976, p<0.001), was significantly associated with TGZ. This is probably because milk production for feeding the family and for generating cash income were the reasons for rearing TGZ. Per capita consumption of milk and meat is also expected to increase following economic growth, urbanization and changing consumer preferences (Herrero *et al.,* 2014).

Fast growth (χ^2=11.242, p<0.001) was significantly associated with TGZ. In all the study areas TGZ was ranked highest with fast growth. Fast growth rate can be of advantage to farmers aiming at producing heifers for sale. The preference for high growth rate of calves might be related to early breeding of heifers, because the farmers are still in the process of building their dairy herds. Besides getting a better price for old dairy cows, farmers receive also a higher price for the bull calves when sold to a fattener because they grow faster (Geuder *et al.,* 2012).

Most of the farmers preferred TGZ because of good mothering ability (χ^2=10.174, p<0.001). The calves usual survive probably because of abundant milk. Hard hooves to withstand muddy places better (χ^2=4.498, p<0.05) was significantly associated with the TGZ. However, one of the farmers visited was cited foot rot in TGZ on following floods in the pasture land.

Breed type preference

More than three quarters (78.8%) of the farmers preferred TGZ. It could have been due to their fast growth rate. This study is in line with (UN, 2017), which states that with the world population expected to reach 9.8 billion in 2050, demand for foods of animal origin is projected to continue increasing. TGZ is still new and was introduced in the country most recently about 9 years ago. As such, because of its genetic potential, it has been much preferred compared to SEAZ. A farmer reported to have exchanged his TGZ bull for two SEAZ heifers.

Table 4: Breed preference

Breed	Frequency	% from total respondents
SEAZ	10	22.2
TGZ	35	77.8
Total	45	100.0

4. Conclusion

Based on the findings of the study, the TGZ was the more preferred cattle breed as it was purported by farmers, to perform better than SEAZ. TGZ is an appropriate genetic mix package for low-input low-output livestock production systems that make up the major part of Uganda's farming systems. This study recommends National Animal Genetic Resources Centre and Databank in Uganda (NAGRC&DB) to put a system in place to follow up the products of the semen given out and train cattle farmers in various aspects of herd management to improve cattle productivity.

5. Acknowledgements

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6. Conflict of interests

The authors declare that they have no conflicting interest.

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A Descriptive Analysis of Breeding Soundness Evaluation Study in Mpwapwa Breed Bulls Based on Established Criteria for the Use of Semen in Artificial Insemination Programmes

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Abstract
The goal of this study was to conduct a descriptive analysis of a BSE study in Mpwapwa breed bulls at TALIRI Mpwapwa research centre based on established criteria for the use of semen in Artificial Insemination (AI) programmes. In the Tanzanian cattle breeding system, breeding soundness evaluation (BSE) is not considered as a common strategic breeding tool that can be used to evaluate bulls' fertility and improve herd reproductive performance and productivity. Despite of being a synthetic dual purpose breed kept for meat and milk production in the arid and semi-arid areas of Tanzania, the potential of Mpwapwa cattle has never been properly realised. The breed had tremendously contributed to the progress of some of the pastoralists and agro-pastoralists in the Tanzanian beef industry, through upgrading of the Tanzania Short horned Zebu ecotypes via crossbreeding. The results from this study indicate that: in most observations, SC for younger bulls were closely similar to SC for older bulls; milk and cream were the highest and lowest recorded colour; and ++ and + were the highest and lowest recorded mass activity/gross motility; 20% and 90% were the highest and lowest recorded individual motility; 50% and 90% were the highest and lowest recorded morphology; and 966 sperm/mL ($x10^6$) and 57 sperm/mL ($x10^6$) were the highest and lowest recorded concentration. Therefore, observed diverse variations particularly in age, SC and semen qualities suggest that Mpwapwa bulls with the best BSE outcomes on SC and semen qualities should be selected for use during the breeding season.

Keywords Mpwapwa breed cattle, Breeding soundness evaluation, Artificial insemination programme and Reproductive performance.

Introduction
The Mpwapwa breed is a synthetic breed, developed from improved Indian dairy breeds (about 60%), African zebu breeds (30%) and European dairy breeds (10%), in order to create a dual-purpose breed, suitable for low to medium production Tanzanian farms. Compared to local Tanzanian zebu cattle, Mpwapwa cattle have higher growth rates and produce four times as much milk, while still having the same level of disease resistance and producing bulls that can be used for draught power (Chawala *et al.*, 2017).

However, the potential of Mpwapwa cattle has never been properly realised. For example, the average milk yield of Mpwapwa cattle is currently no better than it was in the late 1970s (Chawala *et al.,* 2017). The principal driver of this lack of change is that in the 1980s and 90s, structural adjustments to IMF and World Bank policies led to significant reductions in spending in Tanzania on agricultural research as well as privatisation or defunding of parastatal farms, which were crucial to maintaining the development of the Mpwapwa breed. This meant that, genetic development of the Mpwapwa breed was primarily left to semi-commercial smallholder Tanzanian farmers who used natural mating. This reduced selection pressure led to reduced performance of Mpwapwa cattle.

Nonetheless, Mpwapwa breed cattle have contributed to the progress of the Tanzanian beef industry, principally through upgrading of the Tanzania Shorthorn Zebu ecotypes via crossbreeding, with the breed's better performance in terms of growth, disease resilience, and meat characteristics resulting in high demand from pastoralists and agro-pastoralists (Kabuni, 2017).

Thus, increased emphasis on improving the performance of Mpwapwa cattle is likely to have significant benefits for Tanzanian beef production (Kabuni and Laven, 2021). Despite recent changes in policy and government support which mean that selection to improve both productivity and reproductive performance is now feasible (Chawala *et al.,* 2017), significant issues remain, and achieving these selection goals will require a systematic programme of genetic improvement, with cows being mated to the best bulls on both research stations *and* smallholder farms (Chawala *et al.,* 2017). To achieve the latter will require artificial mating and fertility management programmes, both of which are rare on Tanzanian farms.

These programmes will require high quality semen from fertile high genetic merit bulls (Chenoweth *et al.,* 2010). The bulls in the Mpwapwa herd at the Tanzanian Livestock Research Institute (TALIRI) research farm in Mpwapwa are known to have good genetic merit, but we lack good information on their ability to produce high quality semen. The aim of this study was to use a bull breeding soundness evaluation (BSE) to assess the general fertility and semen quality of bulls in the TALIRI Mpwapwa herd, prior to their use in an artificial insemination programme.

Materials and Methods

Study area

TALIRI Mpwapwa is located just outside the town of Mpwapwa in the Dodoma region of Tanzania and is 1000 m above sea level. The two major seasons in the region are dry (May to November) and wet (December to April), with cattle being bred in two breeding seasons: March to May and September to November. The mean daily temperature ranges between 24 and 29°C and annual rainfall is ~720 mm (nearly all of which falls during the rainy season). This study was conducted in February 2021 in the middle of the wet season, prior to the start of the March-May breeding season.

Selection of bulls

All animal-related assessments were approved prior to study start by TALIRI. All bulls that were involved in the study came from the TALIRI Mpwapwa research herd, which in February 2021 had 120 mature (≥24 months) Mpwapwa breed bulls. Bull with a recorded history of previous reproductive problems were excluded (n= 20), and the remaining 100 bulls were then stratified by age (≥24 – ≤28 months, ≥29 – ≤33 months, ≥34 – ≤38 months, ≥39 – ≤43 months, >44 months). Bulls were weighed and body condition scored, and the heaviest 53 bulls were selected for BSE study.

Animal health management and pre-BSE nutrition

After selection, all bulls were weighed, body condition scored, and treated with 5 mg/kg levamisole (Levamizole, Eagle Vet. Tech Co. Kenya). For the five-week period preceding the BSE, bulls were given unrestricted grazing (stocking rate of 0.36 livestock unit/ha) in four 15 ha paddocks. The principal grass species in these paddocks were; *Cenchrus ciliaris, Hyperrhenia rufa, Themeda spp, Cynodon dactylon and*

Chloris gayana. In addition, bulls were trough-fed 0.6 kg/bull/day of a concentrate made from 600 g/kg maize bran, 390 g/kg sunflower seed cake and 10 g/kg salt, and had access to mineral lick blocks (Farmers Centres Ltd, Tanzania) at an allocation rate of 400 g/bull/week. Details of mineral content of the blocks are in supplementary material. No mineral testing was undertaken at any stage of the study, during this period, bulls were dipped weekly in a bath containing 100 g/L of alphacypermethrin (Paranex, Farm base Ltd, Tanzania). Finally, in March 2021, BCS was assessed and weight measured as the bull left the crush after a one month of deworming, supplementation and grazing management.

Allocation to treatment

The 53 selected bulls were grouped based on number order and ear tag number. They were then divided into five BSE Groups (n=10/11) based on that order, with the first bull going to BSE Group 1, the second bull going to BSE Group 2 and so on.

BSE Procedures

Ten bulls were examined per day over a period of five days. All BSE components/tests were conducted based on the procedures outlined by Kastelic and Thundathil (2008):

a) Examination of structural and reproductive soundness (Physical examination)

Bulls were observed while walking on a hard surface. Legs, joints and muscles were checked for lameness and locomotion problems that could affect mating ability, sight was then checked. For examination of reproductive soundness, bulls were confined in a crush and the scrotum and testes palpated, to ensure that they were symmetrical, non-painful and smooth and that the testes were moving freely within the scrotum, similar checks were undertaken for the epididymis and spermatic cords. The prepuce and penis were then checked for hygiene and the absence of abnormalities.

b) Measurement of scrotal circumference (SC)

The testes were then pushed to the bottom of the scrotum and a standard cloth tape placed around the scrotum at its widest point with moderate tension and scrotal circumference (SC) measured.

c) Collection and examination of semen

Semen was then collected by electro ejaculation (Ejakulator, Minitube GmbH, Germany)**.** One person was responsible for inserting the probe into the bull's rectum, one person for regulating the voltage machine and one person for collecting the semen. During the process, the bull was observed for erection and ejaculation, and electrical stimulation ceased immediately upon ejaculation. Semen samples were collected into vials using an ambient manufactured artificial vagina (AV) cone. Semen samples were taken to the laboratory within 3 to 4 minutes of collection.

For semen examination, the collected semen samples were evaluated on the following components:

i. Volume and colour

On arrival at the laboratory, the volume of the collected semen sample was recorded, and its colour assessed. Firstly, the absence of blood and urine staining was confirmed and colour then recorded, semen samples were then placed into a water bath set at 36°C.

ii. Motility

Mass activity/gross motility was assessed using undiluted semen placed on a warm slide without a coverslip. Aliquots of semen were then diluted (1:50µl) in Optixcell diluent (IMV, L'Aigle, France) and individual progressive motility assessed. Both motility measures were assessed using a phase-contrast microscope (MBL2000 Kruss Optronic GmbH, Germany) at x100 magnification.

iii. Morphology

This was assessed using unstained diluted semen specimens (x100 magnification) using a phase-contrast microscope (MBL2000 Kruss Optronic GmbH, Germany). A drop of diluted semen was placed on a glass slide then placed on a microscope stage for examination.

iv. Concentration

The concentration of sperms was measured using spectrophotometer (Accuread Photometer, Biochrom Ltd, USA). A single drop of sodium chloride was drawn and filled in the cavet using a micropipette of 3990 microliter, followed by addition of one drop of raw semen drawn using a micropipette of 10 microliter. The mixture was then thoroughly shaken and placed into spectrophotometer for concentration reading.

Results

Bull age ranged from 28 to 49 months, except for one bull which was 122 months old (median age was 43 months. Median bodyweight at first examination was 230 kg (range 141 -320 kg), at second examination it was 265 kg (range 185 – 365). Median weight gain during pre-breeding 5-week period was 40 kg (range 15 – 72 kg).

There was a moderate correlation between age and body weight at first and second examination (ρ =0.5 and = 0.52 respectively). Removal of the 122 month old outlier resulted in a regression equation of weight at first examination = 1.99 *age (months) + 147.5. The 95% confidence intervals of the slope was 0.81 to 3.17 and for the intercept they were 100.4 – 194.6 (r^2 = 0.19).

All bulls passed the structural soundness and had no detectable abnormalities.

Table 1 Structural and Reproduction Soundness prior to BSE study, Weights and Body Condition Score (BCS) prior and post BSE study during the March-May breeding season 2021

BULL PARTICULARS							
ID	**AGE (months)**	**Structural Soundness**	**Reproduction Examination**	**Body weight at selection (kg)**	**BCS at selection (1-5)**	**Body weight post BSE (kg)**	**BCS post BSE (1-5)**
13117	28	Normal	Normal	230	3	265	4
13119	28	Normal	Normal	183	3	255	4
13122	27	Normal	Normal	170	3	223	4
13128	27	Normal	Normal	225	3	265	4
13139	26	Normal	Normal	163	3	215	4
13174	24	Normal	Normal	190	3	230	4
14566	30	Normal	Normal	205	3	245	4
14574	30	Normal	Normal	230	3	270	4
14590	29	Normal	Normal	185	3	225	4
14718	49	Normal	Normal	190	3	230	4
14722	49	Normal	Normal	239	3	279	4
14739	48	Normal	Normal	264	3	300	4
14747	48	Normal	Normal	314	3	340	4
14750	48	Normal	Normal	268	3	304	4
14762	47	Normal	Normal	242	3	282	4
14785	44	Normal	Normal	285	3	325	4
14791	44	Normal	Normal	265	3	305	4
14802	44	Normal	Normal	246	3	289	4
14804	44	Normal	Normal	238	3	278	4
14817	44	Normal	Normal	276	3	316	4
14835	44	Normal	Normal	200	3	241	4
14836	44	Normal	Normal	244	3	274	4
14841	43	Normal	Normal	238	3	278	4
14842	43	Normal	Normal	220	3	262	4
14843	43	Normal	Normal	235	3	275	4
14852	43	Normal	Normal	273	3	313	4
14858	43	Normal	Normal	240	3	280	4

14865	43	Normal	Normal	220	3	260	4
14869	43	Normal	Normal	252	3	292	4
14887	43	Normal	Normal	195	3	235	4
14889	43	Normal	Normal	232	3	272	4
14891	43	Normal	Normal	215	3	255	4
14901	42	Normal	Normal	230	3	270	4
14903	42	Normal	Normal	203	3	245	4
14911	42	Normal	Normal	222	3	260	4
14922	41	Normal	Normal	205	3	245	4
14924	41	Normal	Normal	217	3	257	4
14952	37	Normal	Normal	230	3	268	4
14965	37	Normal	Normal	215	3	250	4
14966	35	Normal	Normal	185	3	225	4
14967	32	Normal	Normal	187	3	227	4
14975	32	Normal	Normal	190	3	230	4
14976	32	Normal	Normal	240	3	280	4
14977	32	Normal	Normal	250	3	290	4
14980	32	Normal	Normal	204	3	244	4
14983	32	Normal	Normal	208	3	250	4
15946	54	Normal	Normal	220	3	261	4
13161	24	Normal	Normal	212	3	252	4
14755	47	Normal	Normal	141	3	185	4
14770	44	Normal	Normal	235	3	277	4
14916	42	Normal	Normal	230	3	270	4
14925	41	Normal	Normal	320	3	365	4
15195	122	Normal	Normal	285	3	300	4

Table 2 Evaluation of fresh semen samples collected from selected Mpwapwa breed bulls in the BSE study during the March-May breeding season 2021

BULL PARTICULARS			MACROSCOPIC EVALUATION			MICROSCOPIC EVALUATION		Concentration Sperm/mL ($x10^6$)
ID	AGE (months)	SC (cm)	Volume (mL)	Color	Mass activity	Individual motility (%)	Morphology (% normal)	
13117	28	27	4	Milk	++	60	60	132
13119	28	26	2.5	Light Milk	+	60	60	94
13122	27	25	2	Light Milk	+	70	70	125
13128	27	29	8	Light Cream	++	75	75	829
13139	26	25	2	Light Milk	+	80	80	86
13174	24	26	7	Light Milk	+	70	70	117
14566	30	29	5.5	Light Milk	+	70	75	110
14574	30	28	5	Milk	++	55	65	192
14590	29	26	3.4	Light Milk	+	50	50	108
14718	49	28	5	Milk	++	70	75	137
14722	49	30	14.9	Milk	+	70	70	116
14739	48	27	7.5	Milk	++	75	70	223
14747	48	29	3.3	Milk	++	85	90	123
14750	48	28	5	Milk	++	80	85	286
14762	47	28	4.5	Milk	+++	85	90	266
14785	44	28	4	Milk	++	70	80	263
14791	44	27	6.4	Milk	++	75	70	325

14802	44	28	13	Milk	+++	90	90	240
14804	44	26	5.5	Cream	+++	85	75	820
14817	44	28	3	Cream	++	80	80	202
14835	44	25	8	Milk	++	80	80	205
14836	44	28	4.3	Light Milk	+	75	75	102
14841	43	33	6	Milk	++	80	80	746
14842	43	29	8	Milk	+++	85	80	734
14843	43	25	5	Milk	++	80	80	107
14852	43	28	3	Milk	+++	85	87	227
14858	43	27	4	Milk	++	75	75	403
14865	43	28	5	Milk	++	75	85	137
14869	43	27	5.5	Light Milk	++	75	75	103
14887	43	27	7	Light Milk	+	70	80	57
14889	43	27	7.2	Milk	++	75	80	244
14891	43	28	6.4	Milk	++	85	80	253
14901	42	25	3	Cream	+++	90	80	873
14903	42	27	1.9	Milk	++	70	75	100
14911	42	29	8	Milk	+++	90	80	815
14922	41	26	3.2	Milk	++	80	75	310
14924	41	27	3	Milk	++	70	70	305
14952	37	28	5	Milk	+++	80	80	208
14965	37	33	6.5	Milk	+++	90	90	190
14966	35	28	6	Milk	+	20	70	408
14967	32	28	5.8	Milk	+++	75	80	966
14975	32	26	4.1	Milk	++	70	70	326
14976	32	30	8.2	Milk	++	75	75	201
14977	32	26	3.5	Milk	++	70	70	284
14980	32	26	5.5	Milk	+	70	75	315
14983	32	29	2.8	Cream	+++	80	85	896
15946	54	26	10.5	Milk	+	65	80	111
Bulls from which no sperm-rich fraction was collected								
13161	24	26	2.5	Watery				0
14755	47	27	5	Watery				0
14770	44	27	6	Watery				0
14916	42	29	5	Watery				0
14925	41	24	8	Watery				0
15195	122	32	8	Watery				0

Key to mass motility scores	(0) Poor, no waves and the spermatozoa are immobile	(++) Good, less dark waves with moderate movement
	(+) Normal, clear waves with very slight movement	(+++) Very good, lots of dark waves moving rapidly

Semen was successfully collected from 44/50 bulls: only seminal plasma was collected from the remaining 6 bulls.

Discussion

In the Tanzanian cattle breeding system, breeding soundness evaluation (BSE) is not considered as a common strategic breeding tool that can be used to evaluate bulls' fertility and improve herd reproductive performance and productivity. So, the goal of this study was to conduct a descriptive analysis of a BSE study in Mpwapwa breed bulls at TALIRI Mpwapwa research centre based on established criteria for the use of semen in Artificial Insemination (AI) programmes. A general physical examination was conducted

with more attention paid to structural (i.e. legs, joints, muscles and sight), and reproduction (i.e. scrotum, testes, prepuce, penis and sex glands) soundness. There were no evidence of any condition indicating whether bulls had either fertility or structural problems on their reproductive tract and structural soundness. This was contrary to semen examination where some of the evidences concerning volume, colour, motility, morphology and concentration in some of the bulls were observed to be out of the established standards for the use of semen in AI programmes. All bulls' initial body weight had a BCS of 3 which later after a 4 weeks supplementation period improved to BCS of 4 due to increase in their body weight.

Interestingly, bulls' age and SC were inversely proportional to the majority of the bulls. As such, in most observations, SC for younger bulls were closely similar to SC for older bulls (Table 3). As recommended by McGowan *et at.*, (2002), the minimum SC for a 2 year - old *Bos indicus* and its derived crosses in a herd kept under natural grazing system in the aridity areas, should be ≥ 28cm, with less bulls (about < 3%) in a herd failing to meet this recommendation. He also reported that once the bulls attained puberty, SC continues to increase until it reaches the target SC of 34 cm for the 3+ years old bulls. These findings were also reported by Parkinson (2004) and Silva *et al.*, (2014). However, in this study, the youngest bull had 24 months (2 years) with a SC of 26 cm which is 2 cm behind the recommendation, and the oldest bull had 122 months (10.2 years) with a SC of 32 cm which is 2cm behind the targeted SC (34 cm) for the 3+ years old bulls. Among the 53 bulls involved in the study, bulls that were between 2 and 10.2 years old had different SC irrespective to their ages. Since SC is highly heritable, selecting young (i.e. yearling) Mpwapwa breed bulls with the best SC will result into decreased age at attainance of puberty, and improved fertility and reproductive performances (Parkinson 2004; Quirino and Bergmann, 1998).

Table 3. Trend in the relationship between age and SC in some of the older and younger Mpwapwa bulls

Bull ID	AGE (months)	SC (cm)
15946	54	26
14718	49	28
14739	48	27
14762	47	28
14835	44	25
14590	29	26
13119	28	26
13122	27	25
13139	26	25
13174	24	26

However, in some of the older bulls, larger SC were observed (Table 4), but again, the relationship between age and SC in these bulls was not linear.

Table 4. Trend in the relationship between age and SC in older Mpwapwa bulls

Bull ID	AGE (months)	SC (cm)
15195	122	32
14722	49	30
14841	43	33
14965	37	33
14976	32	30
14566	30	29

On the other hand, large variations were observed in the parameters (i.e. volume, colour and concentration) examined for the macroscopic evaluation of the collected semen samples. For example: 14.9 and 1.9 were the highest and lowest recorded volume; milk and cream were the highest and lowest recorded colour; and ++ and + were the highest and lowest recorded mass activity/gross motility. In contrast, slightly variations

were observed for the microscopic evaluation particularly in the individual motility and morphology except for the concentration where large variations were noticed. For example: 20% and 90% were the highest and lowest recorded individual motility; 60% and 90% were the highest and lowest recorded morphology; and 966 sperm/mL ($x10^6$) and 57 sperm/mL ($x10^6$) were the highest and lowest recorded concentration. Regardless of producing reasonable volume, the semen collected from eight bulls were recorded to have watery colouration with no sperm-rich fraction. In addition, some of the older and younger bulls with the highest SC managed to produce semen of acceptable volume (4 - 8 mL), with acceptable percentage of sperm with good motility (40 - 75 %), morphology (65 – 90 %), and concentration (800 – 1500 million/mL), with minimal variations across volume, motility, morphology and diverse variations on concentration (Table 5).

Table 5. Trend in the relationship between age, SC, volume, motility, morphology and concentration for semen samples collected from some older and younger Mpwapwa bulls

Bull ID	Age (months)	SC (cm)	Volume (mL)	Motility		Morphology (%)	Concentration Sperm/mL ($x10^6$)
				Mass activity/Gross motility	Individual motility (%)		
14722	49	30	14.9	+	70	70	116
14841	43	33	6	++	80	80	746
14965	37	33	6.5	+++	90	90	190
14976	32	30	8.2	++	75	75	201
14566	30	29	5.5	+	70	75	110
13174	24	26	7	+	70	70	117

Barth (2000) and Menegassi *et al.,* (2011), reported that larger testicles (SC) are usually observed in bulls from large maturing breeds compared to bulls from small maturing breeds. They pointed out that bulls from the *Bos taurus* breeds (i.e. large maturing breeds) normally have larger testicles (SC) compared to the bulls from the *Bos indicus* breeds. Based on these grounds, there is a linear relationship existing between breed, age, SC and the characteristics of the produced semen. Thus, regardless of the breed, as the age of the bull increases after puberty, the volume and quality of the produced semen also increase as well. However, this relationship ceases once the bull is too old.

Conclusion

Therefore, one of the conclusions that can be drawn on the BSE outcomes of Mpwapwa bulls is that; the observed diverse variations particularly on age, SC and semen qualities suggest that Mpwapwa bulls with the best BSE outcomes on SC and semen qualities should be selected for use during the breeding season. This is because the use of such bulls in a well organised breeding programme can result into production of improved progeny with improved performance, consequently this can lead to improved herd's reproductive performances and productivity. Bulls with poor BSE outcomes on SC and semen qualities on the other hand should be culled from the breeding bulls because their use can result into reduced reproductive performances and productivity.

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Conflict of Interest

Authors do not have any conflict of interest.

Authors Contribution

Authors contributed in the designing, coordination, supervision and implementation of the study. They were also involved in the development of the manuscript.

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Analyzing the Effects of Fuel-wood use on Forest Degradation and Carbon Loss, and Future Policies for Sustainable Energy Utilization in the War-Affected Areas of Ethiopia

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Abstract

The armed conflict in Tigray/Ethiopia has resulted in a total dependence of residents on the scarce forest resources for energy use. Hence, in this study, we monitored fuel-wood transported to three major towns for two seasons and seven consecutive days. The study results showed that fuel-wood transported from the natural forest to each of the studied towns during the war was 16,055 ton per year, which was 628.7% higher compared to the pre-war. The resulting annual average number of trees cut to deliver fuel-wood to each town was 39,563 trees, far beyond the 5429 trees before the war outbreak. Hence, an equivalent of 268-365 hectares of forest at the proximity of each studied town disappeared every year. These in turn resulted to an average annual CO_2 emission of 27692.4 ton compared to 3800.2 ton before the war. To overcome these challenges, a set of five policy recommendations emerged, i.e. issuing fuel-wood permits and regulations; promoting Watershed protection and fuel-wood production approach; improving fuel-wood management and conversion practices; emphasizing on market potential of reforestation activities; encouraging and incentivizing alternative energy sources. It can be concluded that the war and existing siege in Tigray resulted to high forest degradation and carbon loss.

Keywords: fuel wood, forest, carbon, future policy, war affected, season

1. Introduction

Forests constitute important stocks of natural capital by generating benefits including timber, fuel wood, natural erosion regulation, biodiversity and combat climate change (Turner and Daily, 2008; Gibbs and Herold, 2007). They also generate at least 20% of the disposable income of landless and poor families worldwide (Agroforestry-Center, 2011; Schroeder-Wildberg and Carius, 2003), and up to 10% of the gross domestic product of 19 African Nations (World-Agroforestry-Center, 2009). However, these resources are challenged by severe degradation/deforestation (Hosonuma et al., 2012; FAO, 2010; UN-FAO, 2007). Studies showed that Africa's deforestation rate is twice the average in the rest of the world (FAO, 2010). These in turn lead to greenhouse gas emissions (Hosonuma et al., 2012). Taking Ethiopia as an example, the dependency on biomass resulted to an increased CO_2 emission from 5.1 million tons in 2005 to 6.5 million tons in 2010 (Mondal et al., 2018).

Fuel-wood collection and armed conflict represent the major drivers of forest degradation (Dresen et al., 2014; Skutsch et al., 2011). For example, Ethiopia has lost 140,000 hectare natural forest annually in the years between 1990 and 2010, and fuel-wood collection played an important role in the process (FAO, 2010; Haile et al., 2009). Findings also revealed that nearly half of all African countries have recently been

embroiled in civil war (Gleditsch and Salehyan, 2006). In Tigray (northern Ethiopia), the armed conflict which was put in action in November 2020 has resulted in more than 500,000 fatalities, more than 2.2 million people internal displacement, more than 120,000 individuals sexual abuse, and more than 6 million people in need of emergency food aid. All basic services such as banking, telephone, electricity and transport were also immediately suspended after the war broke out (Abay et al., 2022).

While war's adverse effects on human and physical capital have been documented, evidence of its impact on the environment was scant, and the qualitative data collected so far present a mixed story: i) war leads to deforestation; ii) war conserves forest; iii) war has no any relationship with deforestation. These in support of the negative effects of war argued that war creates a power vacuum and breaks down conventional forest management regimes, fostering illegal logging and other resource conflicts (Hoffmann et al., 2018; Castro-Nunez et al., 2017; Fergusson et al., 2014; Allnutt et al., 2013; Glew and Hudson, 2007; McNeely, 2003; Renner, 2002; Kanyamibwa, 1998). A second hypotheses in the opposite direction pointed out that war protects forests by raising the costs of extraction and sale while simultaneously lowering the expected economic returns to farming (Burgess et al., 2015; Richards, 2005; Alvarez, 2003). The third argument revealed no interaction exhibited between armed conflict and forest transitions (Landholm et al., 2019; Ghilardi, 2016; Ordway, 2015; Bensel, 2008). Previous studies also used personal observations, historical accounts, and spatially explicit forest measurements for forest degradation and war relationship assessment (Rudel et al., 2015; Raleigh and Urda, 2007; Draulans and Krunkelsven, 2002). However, the impacts of fuel-wood collection on forest degradation are difficult to quantify even with sophisticated methods (Herold et al., 2011). The valuation of the actual amount of fuel-wood usage and its influence on forest degradation has subsequently been difficult to determine and remained the subject of significant discussion (Mwampamba et al., 2013). Furthermore, the quantity and type of fuel-wood use and associated carbon loss is expected to vary based on seasonal variation and availability of other sources (e.g. cow dung), which calls for further research.

To build a sustainable support to communities, there is a clear need to assess forest degradation for fuel-wood use as well as its environmental risks. Such studies are very important in order to: i) improve our knowledge about the level and impact of fuel-wood consumption and collection on the environment, ii) know the extent to which specific plant communities have been depleted due to wood conversion for fuel-wood purposes. In this study, we monitored fuel-wood transported to towns in Tigray (northern Ethiopia), so as to examine evidence of war's impact on forest degradation and carbon loss. We also collected a set of policy recommendations from series of discussions and review of literatures and policy documents.

2. Materials and Methods

2.1. The Study Area

The study was conducted in three major cities of Tigray (northern Ethiopia), namely: Mekelle (13°29'48"N and 39°28'36"E), Wukro (13°47'32"N and 39°36'13"E) and Maichew (12°47'02"N and 39°32'25"E). These towns are located at the shoulder of the East African Rift Valley, along the Addis Ababa – Asmara main road (Fig.1). The Tigray province belongs to the African drylands (African Sahel), which are often referred to as the *Sudano-Sahelian* Region (BoPED, 1998; Hunting-Technical-Service-Ltd, 1976).

Figure 1. Location of the selected towns in Tigray and Ethiopia

The vegetation cover in the area mostly consist of dense stands of native species like *Juniperus procera*, *Olea africana*, *Erica arborea*, *Dodonaea angustifolia*, *Rumex nervosus*, *Acacia abyssinica*, *Rhus natalensis*, *Euclea schimperi*, *Accacia etibica*, *Dichrostacys cinearea*, *Calpurnea aurea*, *Rhus vulgaris*, *Podocarpus falcatus*, *Hygenia abyssinica*, *Ekebergia spp.*, *Cardia purpurea*, *Mytenus ovatus*, *Salix substrata* and *Balanite egyptica* (BoANR, 1997).

2.2. Method

Data on fuel-wood (dry-wood and charcoal) were collected for two seasons (dry season/January and rainy season/July) and 7 consecutive days in the pre-identified entry-points (Table 1). Data collected include means of transport (e.g. woman, man, donkey and camel), site of origin, type of fuel-wood (charcoal or dry-wood), species and weight of each load.

Table 1: Fuel-wood monitoring locations and their description

Study town	No. of entry points	Description
Wukro	6	Adimeskel, Endaselasie, Megabit 30, High School, Liyu & Atsbi-road
Mekelle	8	Busines, Adishimdhun, St. Mary's Church, Kebelle-18, St.Gebriel Church (kebelle 17), SOS, Arid & Kelkel-Debri
Maichew	10	Bet-Hintset, College, R. Road, St. Michael Church, Bridge, Police Station, Teklehaymanot (kebelle 01), Hareya, Shemeta & Edaga-Kedam

The collected fuel-wood data were organized and classified to analyze the following major components: i) magnitude/amount of fuel-wood transported to each town; ii) conversion of fuel-wood data to the number of live trees; iii) carbon loss due to forest trees degradation; iv) policy recommendations. Baseline information on forest management, for the pre-war, was obtained from key informants' interview and regional/district offices. The weight of each load transported during the monitoring period was summed-up and divided to the number of monitoring days in each season and town. To estimate the amount of fuel-wood transported during the entire year, a conversion factor of 0.75 (for the dry-season) and 0.25 (for rainy-season) was used. The number of months or days in these seasons is different in that the dry-season represents nine months (October to June) and the rainy-season represents three months (July to September) of the year.

To calculate the total weight of fuel-wood transported, the amount of dry-wood used for charcoal making and the dry-wood directly transported are summed-up (Teka et al., 2017). To quantify the amount (weight) of wood required to produce a given amount of charcoal, a conversion factor of 4.7 (Earl, 1983) was applied. In order to estimate the number of trees cut for fuel-wood, the weight/mass of fuel-wood was first converted to volume using a conversion factor of 1.38 (FAO, 1983), and then to live trees using a conversion factor of 0.56 m^3/tree of 20 cm diameter at base height (Dagnoud et al., 1995). The number or density of trees grown in a given area depends on the potential or carrying capacity of the land (Teka et al., 2017). A land with shallow (<50 cm) and degraded soils has a lower potential to support plant growth. However, an equal size of land with a higher depth (>75 cm) and less degraded is able to grow more plants. This assumption was, therefore, taken into account when estimating the area cleared for fuel-wood. Hence, the forest degradation rate in shallow and deep soils, respectively, was estimated using a conversion factor of 44 ton/ha (FAO, 2006) and 60 ton/ha (WBISPP, 1995) (Eq. 1).

$$DF=Q/Kf \text{ --- } (1)$$

where DF = Deforestation (ha/yr), Q = amount of fuel-wood (ton/yr), Kf = conversion factor (ton/ha)

Carbon dioxide (CO_2) emissions was computed using conversion factors for fuel-wood into CO_2 equivalents (Eq.2) based on the Intergovernmental Panel on Climate Change (IPCC), and the 1996 revised report on Guideline for National Greenhouse Gas Inventory (Baral et al., 2019).

$$\text{Carbon dioxide emission } (CO_2e) = \text{biomass of fuel-wood } (ton.year^{-1}) * 0.47 \text{ (carbon)} * 3.67 \text{ } (CO_2 \text{ equivalent}) \text{ ---- } (2)$$

Series of meetings with government officials, individuals (forest experts from GOs, NGOs and wood-fuel traders), and others interested in fuel-wood issues were held. Moreover, we critically reviewed both national and regional policies and proclamations as well as scientific literatures on the issue. These meetings and reviews aimed at: i) explaining the actual fuel-wood management practices in Tigray and the studied towns in particular; ii) identifying possible policy changes to improve upon existing approaches to fuel-wood production.

3. Results

3.1. Forest Management and Fuel-Wood use prior to the War

Between 1997 and 2015, different soil and water conservation practices and technologies (e.g. exclosures, gully rehabilitation, tree plantations and hillside terracing) had been introduced on 510,000 hectares of land in the region/Tigray (Gebrehiwot et al., 2022; Teka, 2019). In the same period, 1307 community watersheds were developed (Teka, 2019). Technically, cutting of native species in these areas is not allowed without special cutting permits from the government (BoANR, 1997). Use of non-forest sources such as electricity and kerosene (Teka et al., 2017); agricultural residues or animal dung and fuel-wood imports from neighboring regions appeared to support fuel-wood demand in the region (Teka et al., 2017; Belay, 2007). Furthermore, demand-side programmes focused on the promotion and dissemination of improved wood-burning stoves; while, supply-side programs focused on establishing fuel-wood plantations, especially in peri-urban and fuel-wood deficit areas. For example, 9687.5 kg day^{-1} of *Eucalyptus camaldulensis* entered to Maichew, 3221 kg day^{-1} to Mekelle and 5200 kg day^{-1} to Wukro town from peri-urban areas just before the war broke-out (BoANR, 2019 unpublished report). As a result, the area of afromontane forest increased from 7.2% in 1986 to 18.6% in 2018 (Gebru et al., 2020).

3.2. Fuel-wood Transported to Each Town

The annual average amount of fuel-wood (both dry-wood and charcoal) transported to each of the studied towns (100,000 to 550,000 population) during the war was estimated at 16,055 tones (Table 2). Except for

Eucalyptus, which was collected from farmers' own woodlot around their homestead, the major point source (more than 55% in Maichew and more than 98% in Mekelle and Wukro) was the natural forest or forest reserve (exclosures) within 40 km radius such as the Desea' forest (in the East).

Table 2: Fuel-wood transported to Markets in 2021/22

Study town	Season 1			Season 2			Estimated Fuel-wood ($Kg.Y^{-1}$)
	Dry-wood ($Kg.D^{-1}$)	Charcoal ($Kg.D^{-1}$)	Total Dry-wood ($Kg.D^{-1}$)	Dry-wood ($Kg.D^{-1}$)	Charcoal ($Kg.D^{-1}$)	Total ($Kg.D^{-1}$)	
Wukro	7,006	2,004	16,425	2,908	517	5,340	4,983,685
Mekelle	59,260	13,279	121,671	15,277	800	19,036	35,044,541
Maichew	22,741	1,093	27,878	5,004	110	5,519	8,135,21**6**
Average	**29,669**	**5,459**	**55,325**	**7,730**	**476**	**9,965**	**16,054,481**

Kg = kilogram, D = Day, Y = Year

The number of tree species harvested for dry-wood collection varied between 3 and 19 (Fig.2). In **Maichew town**, only 7 out of the 17 species covered 89% of the incoming dry-wood. The dominant species were, *Eucalyptus camaldulensis* (43%) > *Erica arborea* (19%) > *Dodonaea angustifolia (*9%) > *Rumex nervosus* (8%) > *Acacia abyssinica (*4%) > *Rhus natalensis/Euclea schimperi* (3% each). In **Mekelle city**, only 3 out of the 13 species covered 94% of the incoming dry-wood. These species were represented by *Accacia etibica* (65%) > *Olea europea* (26%) > *Rhus natalensis* (3%). In **Wukro town**, only 2 out of the 3 species (68.6% *Olea europea* and 31.2% *Accacia etibica*) covered 99.8% of the incoming dry-wood.

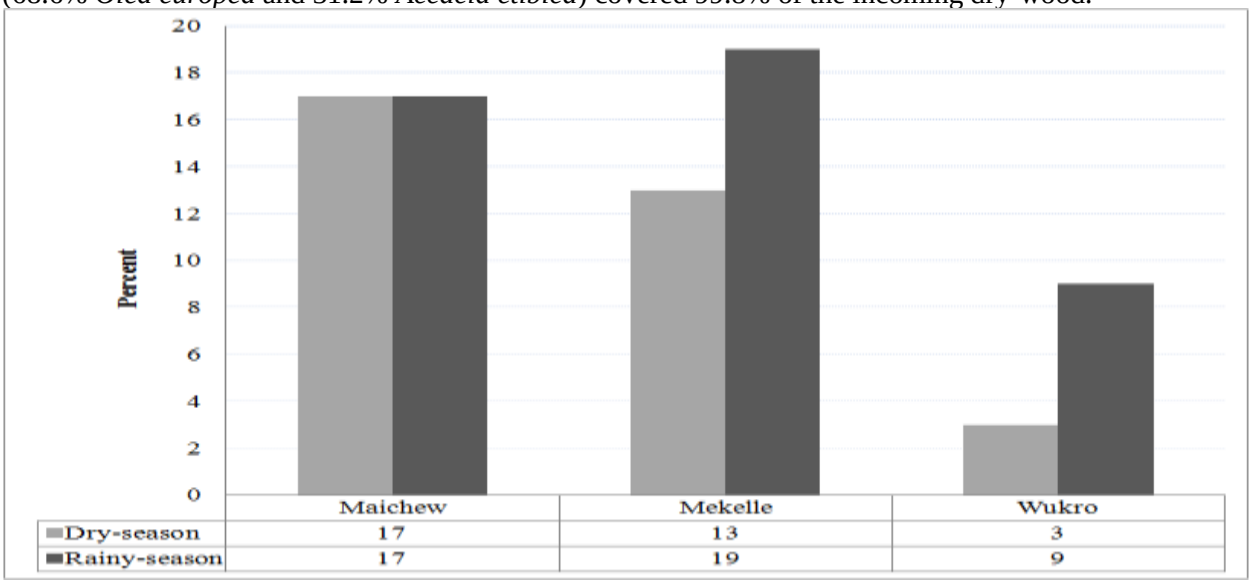

Figure 2. Number of tree species used for dry-wood collection

3.3. Forest Degradation for fuel-wood use

The annual average number of trees cut to deliver fuel-wood to each town before the war was estimated at 39,563, which was much higher than the 5429 trees loss just before the war outbreak. This in turn resulted to disappearance of an equivalent of 268 to 365 hectares of forest annually at the proximity to each studied town (Table 3). Majority of the respondents/farmers' cut live trees 2 or 3 times a week (depending on the occurrence of holidays) and hide them under bushy/denser forests for 2 or more weeks or transport them to nearby villages/houses in order to make them dry.

Table 3: Forest trees harvested for fuel-wood use and transported to the surveyed towns

Study town	Estimated Fuel-wood (ton.Year^{-1})	Volume (m^3.Year^{-1})	No. of trees harvested per year	Deforestation rate (ha.year^{-1})	
				Soils < 50 cm deep	Soils >75 cm deep
Wukro	4,983.7	6,877.5	12,281	113.3	83.1
Mekelle	35,044.5	48,361.5	86,360	796.5	584.1
Maichew	8,135.2	11,226.6	20,047	184.9	135.6
Average	**16,054.5**	**22,155.2**	**39,563**	**364.9**	**267.6**

3.4. CO_2 emissions for fuel-wood use

The illegal tree cutting for fuel-wood resulted in CO_2 – emission of 27,692.4 ton.year^{-1} (Table 4), which is much higher than the 3800.2 ton.year^{-1} just before the war.

Table 4. Carbon loss for fuel-wood use in each surveyed town

Study town	Estimated Fuel-wood (ton.Year^{-1})	CO_2 – emission (ton.Year^{-1})
Wukro	4,983.7	8,596.4
Mekelle	35,044.5	60,448.3
Maichew	8,135.2	14,032.4
Average	**16,054.5**	**27,692.4**

3.5. Identified Policy Recommendations

To overcome the challenges of continuing forest degradation, a set of five future policy recommendations, which have relevance for fuel-wood management in Tigray and the developing world emerged: i) Issuing fuel-wood permits and regulations; ii) Promoting watershed protection and fuel-wood production; iii) Improving fuel-wood management and conversion practices; iv) Emphasizing on market potential of reforestation activities; v) Encouraging and incentivizing alternative energy sources.

4. Discussion

Tens of thousands of hectares of uplands in Tigray have been designated as 'critical watersheds' since 1997, and efforts were underway to replace what are perceived as inappropriate land-use practices with ecologically appropriate reforestation projects. Moreover, to reduce the dependence of energy on natural forest, different non-forest sources were introduced. For example, fuel-wood use from *Eucalyptus* plantations outside the forest reserve prior to the war amount to an annual average of 2203.7 tons per town (BoANR, 2019 unpublished report). This corresponds with the findings in other developing regions that stated trees outside forests meet the bulk of local fuel-wood requirements (Koopmans, 2005; Bhattarai, 2001). Our findings also support the survey results in other developing countries that revealed residents were often responding to forest product scarcities through increased on-farm tree plantation (Rudel et al., 2005). The overall result was a threefold increase, from 7.2% in 1986 to 18.6% in 2018, in the area of afromontane forest (Gebru et al., 2020).

The annual average amount of fuel-wood transported to each of the studied town, during the war, was 628.7% higher than the amount transported just before the war outbreak. This is much higher than the estimated amount for Goma city of Democratic Republic of Congo (about half million population) that consumed an estimated 2,947 tons of charcoal and 3,926 cubic meters of wood (Emily, 2010). Unlike to the pre- war outbreak, the major fuel-wood source during the war was the natural forest or forest reserve (exclosures). This also corresponds with the findings for South-Eastern Ethiopia that revealed about 83% of the respondents collected fuel-wood from forest reserves (Mohammed et al., 2020).

As stated in Figure 2, the number of tree species harvested for dry-wood collection varied between 3 and 19 depending on the agro-ecology and proximity to the natural forest. These findings are in par with the household level common species consumption study results for urban areas of Tigray including the study

districts (Teka et al., 2017). These authors revealed that *Olea europaea* (68.6%) in Wukro town, *Acacia etibica* (51.5%) in Mekelle city and *Eucalyptus camaldulensis* (40.8%) in Maichew town were the dominant species used for fuel-wood. Other studies also found out that the most exploited tree species in the remnant dry afromontane forests were *Juniperus procera*, *Olea europea*, *Podocarpus falcatus* and *Hygenia abyssinica* (Mohammed et al., 2020; Zenebe et al., 2020).

The war in Tigray has also resulted to an annual loss of extra 5429 trees as compared to the pre-war. In line to our findings, estimates for the Virunga National Park of Goma city in Democratic Republic of Congo revealed that at least 432 to 576 hectares of tropical forest disappear every month (Emily, 2010). The tree harvesting method was total felling/total destruction. A similar result was also reported for South-Eastern Ethiopia and other developing countries that revealed more than 83% of the respondents collected fuel-wood by cutting live trees (Mohammed et al., 2020; Asian-Development-Bank, 1995).

The illegal tree cutting for fuel-wood, in the study area during the war, resulted in a 3800.2 ton.year^{-1} higher CO_2 – emission compared to the pre-war. This corresponds with the interview results from 217 households in Kankali community managed forest of Nepal that revealed 60% of the households who depend on fuel-wood for cooking apparently emit approximately 13.68 tons of carbon dioxide annually (Baral et al., 2019). Other studies also showed that total distraction of a single tropical tree can lead to an annual loss of 22.6 kg or 2.5–3.6 ton.ha^{-1} of carbon dioxide (World-Agroforestry-Center, 2011).

Hence, the following five sets of policy recommendations were identified, from experts' judgment and review documents, to overcome the challenges of continuing forest degradation:

i) **Issuing fuel-wood permits and regulations:** producing, packaging, transporting and trading fuel-wood and charcoal can employ large number of people and earn revenues for the government or other resource owners. For example, in Dareselaam and Malawi, the fuel-wood and charcoal trade involves 125,000 and 92,800 people, respectively (World-Agroforestry-Center, 2009). Hence, issuing fuel-wood permits and regulations can play a pivotal role in the sustainable utilization of forest resources. The most commonly used forest management regulations worldwide are: private land-owners can cut planted trees on their own land without a permit, but they must apply for a transport permit if they want to sell any of the wood from these trees somewhere else; special cutting and transport permits are required if a landowner wants to cut naturally growing tree species found on his/her lands (Bensel, 2008); there should be a binding rule for permitted or legal fuel-wood suppliers (Parviainen et al., 2009); a certified site must have a minimum of 5–10 trees per hectare (Parviainen et al., 2009); where stump extraction is permitted, at least 30% on stumps shall not be harvested (Kankaanpaa et al., 2005); for ensuring biodiversity, at least 5 m^3.ha^{-1} of dead wood shall be left on the regeneration site (Kankaanpaa et al., 2005); the landless should be given seedlings provided free of charge and given ownership of trees in return for their guarding and watering the seedlings (Bensel, 2008).

ii) **Promoting watershed protection and fuel-wood production:** many of the towns in Tigray region are laid in a location surrounded by mountainous topography which is exposed to flooding (Teka et al., 2017). To minimize the effects of flooding and at the same time fulfilling the fuel-wood demand of their population, realization of urban watershed is a proved approach (Parviainen et al., 2009). The "analysis of household's energy consumption, forest degradation and plantation requirements" study for the eastern Tigray in 2017 recommended an estimated annual tree plantation of area ranging from 286 ha around Maichew town to 21,684 ha around Mekelle city (Teka et al., 2017). Hence, planting suitable tree species on cultivated lands, strip lands (e.g. farm bunds and boundaries, roads, canals and drains), degraded forest lands and uncultivated degraded lands (non-forest and non-cultivated lands) can help supply fuel-wood to the urban poor (Saxena, 1997). For example, half of the fire wood burned in Thailand, more than three-quarters in Indonosia, Java, Pakistan & Vietnam, and four-fifth in India is cut from farm land and other non-forest areas (World-Agroforestry-Center, 2011). For household usage, families should be encouraged

to plant a mix of fast growing and multiuse trees on their homesteads so as to diversify income sources, overcome potential problems from pest or disease infestations and increase biodiversity in addition to securing their energy sources (Bensel, 2008).

iii) **Improving fuel-wood management and conversion practices:** fuel-wood burning in many developing countries including the study area is done on less efficient energy conversion traditional stoves (Manaye et al., 2020; Dresen et al., 2014; Gulilat et al., 2011; Shanko and Lakew, 2011; Bensel, 2008; Feleke, 2007). The energy conversion efficiency of these stoves ranges between 10.2 and 19% (Bhattacharya and Salam, 2002) compared to the 40% fuel-wood savings by improved cooking stoves (Manaye et al., 2020; Dresen et al., 2014). These in turn led to a total GHG emission of about 110 g of CO_2 equivalent per mega joule of useful energy compared with 42 g for improved wood-stoves and 5 g for biogas (Bhattacharya and Salam, 2002). Hence, application of fuel-wood use saving mechanisms can help reduce the burden on natural forest and thereby reduce carbon emission. The use of improved cooking stoves results in 0.65 tons of carbon dioxide equivalents (CO_2e) emission reduction per stove per year (Manaye et al., 2020).

iv) **Encouraging and incentivizing alternative energy sources:** any sustainable development solution in the household energy sub-sector must focus on renewable energy sources (e.g. Biogas, Bio-fuels, Wind, Solar and Hydro-power). For instance, **Biogas** stoves have conversion efficiency of 55% (Bhattacharya and Salam, 2002). A biogas technology, having 8 m^3 digester size, can save about 1423 kg of fuel-wood with an emission reduction potential of 2.1 tons of CO_2e per biogas plant (Desta et al., 2020; Dresen et al., 2014). Growing **Bio-fuels** (e.g. Jatropha and sweet sorghum) can also help alleviate energy shortages, sequester carbon and improve incomes (World-Agroforestry-Center, 2009). **Wood residues** generated during harvesting and processing is another alternative energy source. For example, wood residues fulfilled the Cameroon's total electricity demand of 3,320 Gigawat-hours, 60% of the electricity consumed in Gabon and 12% in Nigeria (World-Agroforestry-Center, 2009). Similarly, **energy** from hydro-, solar or wind power is a good substitute for fuel-wood that is harvested unsustainably (Owusu and Asumadu-Sarkodie, 2016; Kaunda et al., 2012; Panwar et al., 2011).

v) **Emphasizing on market potential of reforestation activities:** Many reforestation and conservation projects continue to promote tree planting primarily for ecological reasons (Murray and Bannister, 2004). However, studies suggested that the most promising approach is enabling local communities and farmers to rehabilitate and manage forest lands and directly benefit from their efforts (Chokkalingam et al., 2006). These can be achieved if and only if they provide market opportunities. Farmers and landowners are more likely to undertake tree planting and management when these are perceived to provide market opportunities (Murray and Bannister, 2004; Russell and Franzel, 2004; Scherr, 2004).

5. Conclusions

In view of the limited access to electricity and kerosene, due to the existing war and siege, most households in Tigray excessively depend on fuel-wood. The study results indicated that the annual average amount of fuel-wood transported to each of the studied towns was 16,055 tones, which is by 628.7% higher than the amount transported just before the war outbreak. The number of tree species cut for the purpose ranges from 3 in Wukro town to 19 in Mekelle city. The resulting annual average number of trees cut to deliver fuel-wood to each town was estimated at 39,563 trees, which was much higher than the 5429 trees loss just before the war. Hence, an equivalent amount of 268 to 365 hectares of forest at the proximity to each studied town disappeared every year. These in turn resulted in an average CO_2 emission of 27692.4 $ton.year^{-1}$, and a seven fold increase compared to the situation before the war outbreak. It can, therefore, be concluded that the high dependency of the community on Fuel-wood resulted to a high forest degradation and carbon loss. Hence, the following policy recommendations, which have relevance for fuel-wood management in Tigray and the developing world, are suggested: i) Issuing fuel-wood permits and regulations; ii) Promoting

watershed protection and fuel-wood production; iii) Improving fuel-wood management and conversion practices; iv) Emphasizing on market potential of reforestation activities; v) Encouraging and incentivizing alternative energy sources.

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Application of GIS in the Study of Spatial Variability of Selected Soil Properties in Southeastern Nigeria

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Abstract
Assessment of spatial variability of soil is a key component to sustainable food production. Soil sampling is being threaten by the insecurity ravaging the African nations. The combination of both the conventional analytical method and geostatistical method were used to analyse the variability of some soil properties in three state of the Southeastern Nigeria. A total of 18 profile pits were dug and sampled on soils underlain by six different parent materials. The sand content, clay content, porosity, pH, organic carbon (OC), available phosphorus (AP), base saturation (BS) and effective cation exchangeable capacity (ECEC) were analysed using standard analytical procedures. The study reveal that sand content predominate other soil properties with highest value of 87.7%. The clay content varied and falls within the range of 9.31 – 3.45%. The total porosity was more than 40% in all the soils studied. The pH ranged from 5.47 – 7.14, the OC ranged from 3.9 – 11.7g/kg, the AP ranged from 7.21 – 12.45mg/kg, the BS ranged from 37.75-89.13% and the ECEC ranged from 4.03 – 11.23cmol/kg. The result of the geostatistical analyses revealed that spatial dependence (SD) below 25% indicating strong SD was observed in porosity, organic carbon, phosphorus and ECEC. Moderate SD was observed in in sand content, clay content and pH while weak SD was observed in the percentage base saturation. The SD was apparent in 13873 -79156.76m ranges. The coefficient of determination (R^2) of clay content and ECEC were greater than 0.5 indicating a good fit. The Root Mean Standardized Error (RMSE) <1 was observed in pH and ECEC indicating good prediction of the measured value. This study suggest that OK interpolation method can be used to predict spatial distribution of some soil properties at large scale with few sampling points.
Keywords: spatial variability, soil properties, spatial dependency, ordinary kriging.

Introduction
The use of current technology to achieve food production is critical at this moment most Africa countries are faced with security challenges. The drastic increase in global population has brought about an increasing demand of agricultural production. Therefore, there is urgent need to outline strategies that will encourage sustainable production of food with consideration to soil nutrient management (Jones, Pual, and Wither, 2013). It is not only enough to understand the nature, properties, dynamics and functions of the soil as part of landscapes and ecosystems but also how soil information are managed for effective use. Following the rise in the use of technology, many innovations have been put in place to sustain human existence on earth. Geographic Information System, a very efficient computer-based tool is playing an important role in soil resource management. It is a most recent tool capable of storing, retrieving and analysing different types of soil data for proper management of soil resources (Igboekwe and Akankpo, 2011).

The accessibility and availability of geographic information systems (GIS), remotely sensed spectral data, global positioning systems (GPS), digital elevation models (DEM), Interpolation models and software have made significant contributions to the science and art of soil surveying (U.S.D.A, 2019).

Several studies (Hengl, Rossiter, and Husnjak, 2003; Nkwunonwo and Okeke, 2013) have demonstrated that such activities are effective methods of assuring the continuous availability of soil data.Conventional data displaying systems like the use of tables as seen in most research work in Southeastern Nigeria is becoming obsolete since the increase in the use of technology. Setia, Verma, and Sharma (2012) used a decision support that is based on GIS system to determine the potentials and constraints of different types of soils for crop production. The aim of this research is to monitor the spatial variability of some properties in large scale with few sampling points in order to encourage precision farming especially in areas facing security challenges in Africa.

Materials and Methods

Location

The study was carried out in three states in the Southeast geopolitical zone of Nigeria. Southeastern Nigeria is located within latitudes 4° 47' 35'' N and 7° 7' 44'' N and longitudes 7° 54' 26'' E and 8° 27' 10'' E. The three states studied include Anambra, Enugu and Ebonyi State (Fig 1). The geomorphology is predominantly covered by laterite soil. The average annual rainfall and temperature is 1600mm and 26^0C. Southeastern Nigeria is located in the rainforest natural vegetation belt. This vegetative zone is characterized by evergreen trees toward the south and rainfall-savanna forest toward the north. The rainfall-savanna forest is made up of trees and grasses scattered everywhere. Anthropogenic activities have significantly impacted this vegetation with variety of plant types like cassava (Manihotspp), plantain (Musa spp), oil palm (Eleais guineesis), maize (Zea mays), yam (Dioscorea species), ogbono tree (Irvingia gabonensis), ube (Dacryodes edulis), mango (Mangifera spp.) etc. About 50% of the total area is used as cultivated land.

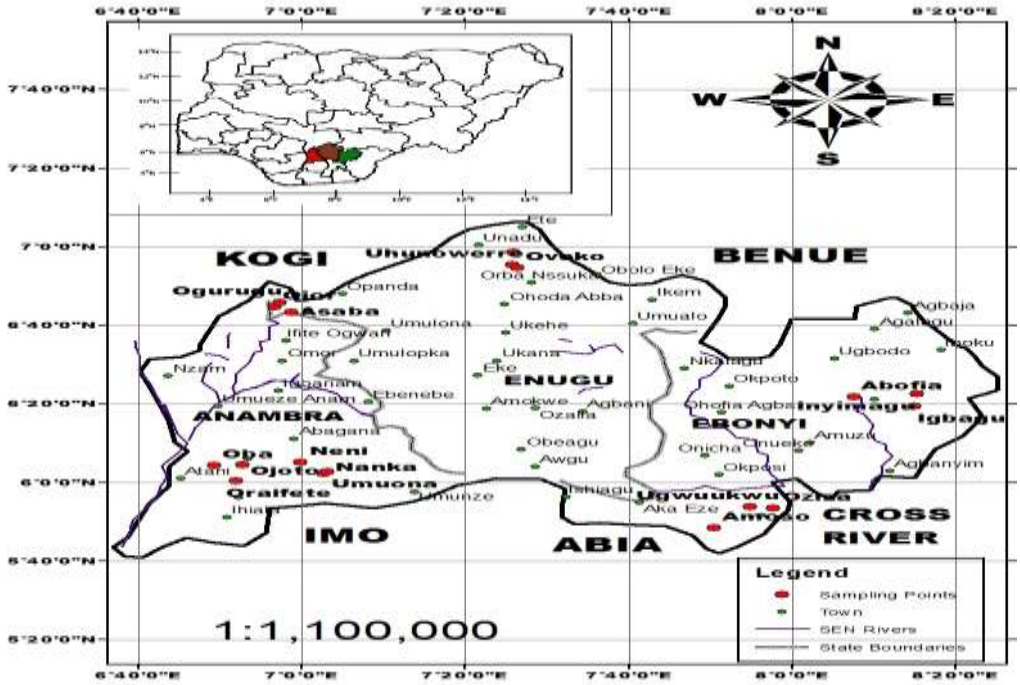


Figure 1: Location Map of the Study Area indicating its Position in the Map of Nigeria

Soil Sampling and Measurements

A reconnaissance field study was carried out for the purpose of identifying the parent materials and free survey approach (Onweremadu, Opoke and Ohaeri.,2014) was used in locating sampling points with the help of a geology map. A total of six parent materials were identified and three profile pits were dug on each of them making a total of 18 profile pits. The parent materials identified were Imo clay shale in Ojor, Asaba and Ogurugu (Enugu State), Ajali sandstone in Uhunowere, Ovoko and Enugu Ezike (Enugu State), Asu river group in Inyimagu, Igbeagu and Abofia (Ebonyi State), Afikpo Sandstone in Ugwukwu, Oziza and Amoso (Ebonyi State), Ogwashi-Asaba formation in Oba, Oraifite and Ojoto (Anambra State), and Bende-Ameki formation in Neni, Umuona and Nanka (Anambra State). Mapping units were delineated and soil profile were located at undisturbed and slightly disturbed site. Soil profiles were described according to FAO guidelines for soil description (FAO, 2006) and sampled according to the genetic horizons in December 2019 and January 2020. Soil samples were subjected to various physical and chemical analyses. Standard analytical techniques were used to determine the soil particles (Hydrometer method; Gee and Or, 2002), porosity (Vomocil, 1965), pH (1:1 soil/water ratio using digital pH meter), soil organic carbon (SOC) (Walkley and Black dichromate wet oxidation method; Nelson and Sommers,1982), phosphorus (P) (Bray II method; Olsen and Sommers 1982), The exchangeable bases (extracted with 1N NH4OAc solution) and exchangeable acidity (leached out from the soil with 1N KCl) were determined and the results were used to calculate the Effective Cation Exchange Capacity (ECEC) and the base saturation (BS). The mean result from each profile was used for the geostatistical analysis.

Geostatistical Methods

All sites were geo-referenced using handheld GPS receiver. The shape files were developed from the open street map and the coordinate of each location from the GPS was used to place the sampling sites in the actual geospatial position using Arc GIS 10.2 software. The soil properties determined were linked to the respective positions in the GIS database for the purpose of generating attribute maps. The geostatistical analyses were carried out with the help of the Geostatistical Wizard of Arc GIS 10.2 in the geostatistical analyst extension. The normality of the spatial distribution of the soil properties was monitored using descriptive statistics. Soil data that are not normally distributed was transformed using log transformation for easy interpolation. The spatial distribution structure of the soil properties analysed was monitored using semivariogram and this was based on the regionalized variable theory and intrinsic hypotheses (Nielsen and Wendroth 2003). The spatial dependence is strong if SD < 25%, moderate for SD between 26% and 75% and weak with SD > 75% according to method detailed by Cambardella et al., (1994). The GIS software was also use to perform the ordinary kriging (OK) interpolation for the prediction of the values of the un sampled locations (Wang, 2018). The best-fit model with the lowest value of residual sum of squares was selected for each of soil property analysed. Cross-validation technique was adopted for evaluating and comparing the performance of Ordinary Kriging interpolation method. The cross validation techniques revealed the root mean square error (RMSE) and the coefficient of determination (R^2).

Result and Discussion

The results of the selected soil properties are presented in Table 1. The sand content of the soil predominates other soil particles. This is in line with the study of Onweremadu et al., (2011) and, Okebalama, Igwe and Okolo., (2017) on soils of Southeastern Nigeria. The highest value for sand content (87.7%)was observed in soils of Ogwashi-Asaba Formation (OAF) while the least sand content (47.05%) was observed in the soils of Imo Clay Shale (ICS). The clay content of the soils varied. The soils of Asu River Group (ARG) had the highest clay content (25.29-31.45%) while the soils of Ogwashi-Asaba Formation (OAF) had the least clay content (9.31- 15.48%). This variation may be as a result of the varying underlying parent materials. The total porosity (Table 1) of the soils studied were between the range of 40 -50.16g/kg which is very suitable for crop production. Landon (1991) noted that total porosity of soils less than 40% is likely to restrict root growth due to excessive strength. The pH values of the soils studied were generally acidic

irrespective of their parent materials. The least pH value (5.47) was observed in one of the soil of Bende-Ameki Formation (BAF) while the highest pH value (7.14) was observed in one of the soil ARG (Table 1). However, according to Landon (1991) pH range of 5.50 to 7.50 provides the most satisfactory plant nutrient levels for most crops and most of the soils studied fall within this range.

Table 1: Physical and Chemical Properties of Soils

Sampling Location	Sand (%)	Clay (%)	Porosity (%)	pH	OC (g/kg)	Phos (mg/kg)	BS (%)	ECEC cmol/kg
Imo Clay Shale								
Asaba	47.05	26.98	46.35	6.07	8.65	9.14	57.65	8.94
Ojor	76.72	15.00	45.25	6.46	6.04	7.50	83.99	7.48
Ogurugu	602.80	19.87	47.17	6.24	7.07	6.84	62.34	6.68
Ajali Sadstone								
Enugu Ezike	73.12	22.92	44.77	6.63	8.16	7.64	77.90	4.87
Uhunoowerre	81.77	14.20	48.53	6.69	6.00	8.35	72.20	5.10
Ovoko	80.12	17.00	46.00	6.46	6.44	7.32	89.13	5.43
Asu River Group								
Inyimagu	53.32	25.20	42.33	6.04	6.52	11.91	37.75	8.99
Igbagu	51.52	28.87	46.32	7.14	11.73	8.28	64.87	11.23
Abofia	48.52	31.45	40.70	6.15	10.90	12.43	61.23	9.12
Afikpo Sandstone								
Ugwuukwu	76.52	15.70	43.89	6.06	5.50	9.40	57.43	6.75
Oziza	62.92	25.92	42.92	5.62	8.16	7.39	54.85	8.32
Amoso	77.52	13.57	45.56	6.48	2.64	6.96	61.39	6.01
Ogwashi-Asaba formation								
Oba	87.72	10.12	48.66	5.59	5.76	8.08	71.40	5.57
Oraifete	82.00	15.48	44.85	5.92	5.72	7.83	64.78	4.59
Ojoto	87.13	9.31	48.06	5.59	3.90	7.14	71.36	4.65
Bende- Ameki formation								
Neni	82.32	14.37	47.10	5.86	5.14	7.21	72.41	4.88
Umuona	66.96	26.88	49.71	5.83	8.10	7.27	60.15	4.03
Nanka	78.72	18.00	50.16	5.47	6.24	7.36	55.64	4.53

The soils of ARG had the highest organic carbon content value of 6.5-11.7g/kg while the soils of OAF had the least organic matter content value of 3.9-5.8g/kg. Soil with greater clay content typically have greater soil organic carbon stocks due to physical protection of organic minerals. The available phosphorus in most of the soils studied was generally below 10ppm/g and thus classified as low according to Landon (1991) nutrient requirement for most crops. The highest value of phosphorus was recorded in soils of ARG with range of 8.28 – 12.43mg/kg while the least value was observed in soils BAF with range of 7.21 – 7.36mg/kg. The low available phosphorus observed could be due to the fact that in the humid tropics like Southeastern Nigeria, there is rapid fixation of phosphorus compound found in the soil into unavailable form by forming complexes with iron, aluminium and manganese (Brandy and Weil, 2002, Onweremadu and Dunigbo, 2007).

Table 2: Semivariogram and Cross Validation Parameters.

Soil Prop.	Model	Nugget	Sill	Range (m)	Spatial Ratio (%)	Spatial Class	R^2	RMSE
% Sand	Gaussian	0.0156	0.0526	79156.76	29.66	moderate	0.5412	9.5452
% Clay	Spherical	0.0444	0.135	25055.66	32.89	moderate	0.256	6.0181
Porosity	Spherical	0.0003	0.0025	15557.59	12	strong	0.4242	2.0665
pH	Gaussian	0.0021	0.007	145385	30	moderate	0.3291	0.398
OC	Spherical	0.007	0.1298	18553.26	5.39	strong	0.0684	2.3749
Phosphorus	Spherical	0.3174	1.6654	14143	19.06	strong	0.0914	1.2716
ECEC	Gaussian	0.011	0.1162	13873	9.47	strong	0.7241	0.9543
BS%	Gaussian	0.023	0.0242	68217.26	95.04	weak	0.1325	11.78

Generally, the base saturation ranges from 37.75% to 89.13% across the study sites. The effective cation exchangeable capacity (ECEC) of the soils studied was generally low with range of 4.03 – 11.23 $cmolkg^{-1}$. Soils of South-eastern Nigeria had earlier been reported to have low ECEC and basic cations (Ahukaemere et al., 2016). The low effective cation exchange capacity of these soils revealed low capacity of these soils to retain nutrient elements in the soil exchange site (Chikezie et al., 2010).

Geostatistical Analysis

Table 2 shows the results of the semivariogram parameters (range, nugget, and sill) for each soil property studied with the best-fitted model (Fig. 2A and Fig. 2B). The degree of spatial variability of these soil properties are affected by both structural (climate, parent materials, topography and other natural factors) and stochastic factors (fertilization, cropping system and other anthropogenic activities). The lower spatial ratio (Spatial dependency) suggests that structural factors plays significant role in spatial variability while higher value suggest that that stochastic factor primarily caused spatial variability (Shit, Bhunia and Maiti, 2016).

The spatial dependence (SD) was graded as strong if SD < 25%, moderate for SD between 26% and 75% and weak with SD > 75% (Cambardella et al, 1994). The SD below 25% indicating strong spatial dependence was observed in porosity, organic carbon, phosphorus and ECEC. Moderate spatial dependency was observed in in sand content, clay content and pH while weak spatial dependence was observed in the percentage base saturation. The spatial dependence was apparent in 13873 -79156.76m ranges (Table 2).

The semivariance function model fits Gaussian curve for sand content, pH, ECEC and percentage base saturation. The Spherical curve was the best fit model for clay content, porosity, organic carbon and phosphorus. The curves gradually increase with increasing spatial distance before stabilizing (Fig. 2A and Fig. 2B). All the soil properties studied have coefficient of determination (R^2) values of 0.0684 – 0.7241. The R^2 measures the goodness of fit (Harisuseno et al 2020; Ikuemonisan, et al., 2020). The R^2 of clay content and ECEC were greater than 0.5 indicating a good fit (Table 2).

The Root Mean Standardized Error (RMSE) indicate how closely the model predicts the measured values. The lower the RMSE, the better the prediction. RMSE <1 was observed in pH and ECEC. The highest RMSE was observed in percentage base saturation (Table 2).

Figure 2A: Semivariogram Curves

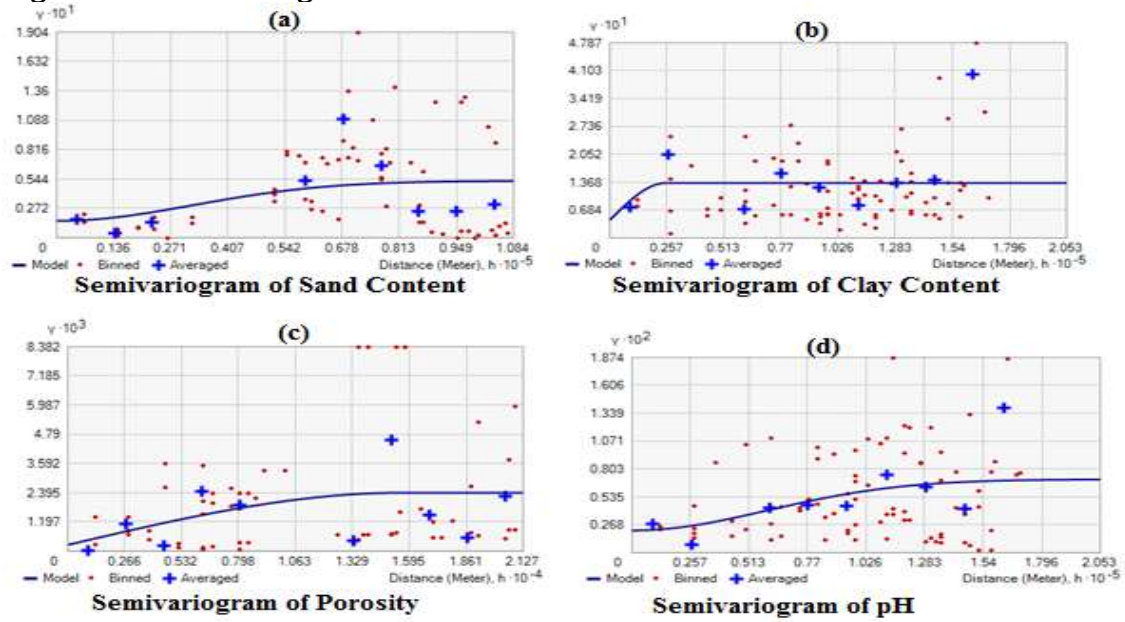
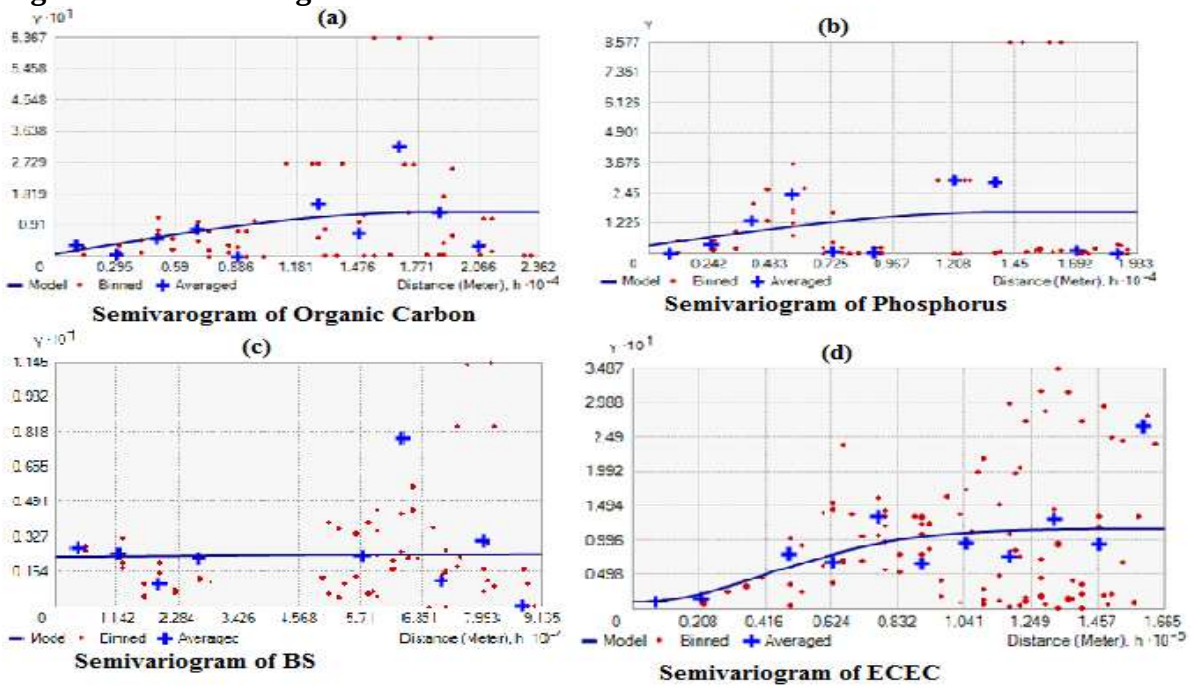


Figure 2B: Semivariogram curves



Spatial Distribution of Soil Properties

The spatial distribution of soil properties is displayed in Figure 3 to 10. The sand content (Fig 3) followed an observable pattern with the highest percentage of it found in the southwest part of the study area in Atani, Anambra State and it reduced towards the eastern part of the study area in Ebonyi State. Asu river group which covered more than 80% of the soils of Ebonyi State had less sand content when compared to other

soils studied. The sand content of 69.68 -76.55% and 60.18 to 69.68% covered the entire soils of Enugu State. The highest clay content was found in the soils of Ebonyi State and the least clay content was found in soils of Anambra State. The clay content of 15.42-23% covered most soils of Enugu State.

The lowest porosity was observed in soils around Ezzagu and Abofia in Ebonyi state. The porosity of 44.91-47.11% covered most of the soils of Enugu State. The least porosity was also observed in Nanka in Anambra State and this could be attributed to the sandy nature of these soils. Porosity also had spatial ratio less than 25%, and this implies that structural factors such as parent materials, topography and other natural factors plays significant role in its variability. The pH of the soils studied followed an observable pattern (Figure 4) with the soils in southern part Anambra having the lowest pH and soils in the northern part of Enugu and Ebonyi State having the highest pH. The variability in the degree of leaching and the dominant clay mineralogy must have contributed in the variation of pH.

Organic carbon content of 6.38-7.57 gkg^{-1} and 5.54- 6.38 gkg^{-1} covered most of the soils of Enugu State while organic carbon content of 5.54- 6.38 gkg^{-1} and 4.35-5.54 gkg^{-1} covered most soils of Anambra State. Greater percentage of organic carbon was observed in soils of Ezzagu and Igbeagu of Ebonyi State and organic carbon of 7.57-9.28gkg^{-1} covered almost the rest of Ebonyi State. Available phosphorus (Figure 5) was also observed to be higher in Agbaja, Agalagu, Ugbodo, Abofia and Ezzagu all in Ebonyi State. The least value of 6.84-8.25$mgkg^{-1}$ covered the entire Anambra State.

The value of 8.25. 9.79 $mgkg^{-1}$ and 6.84-8.25$mgkg^{-1}$ covered most of soils of Enugu State. All these observations revealed that pH affected the availability of phosphorus because there is rapid fixation of phosphorus compound in acidic soil into unavailable form by forming complexes with iron, aluminium and manganese (Brandy and Weil, 2002).

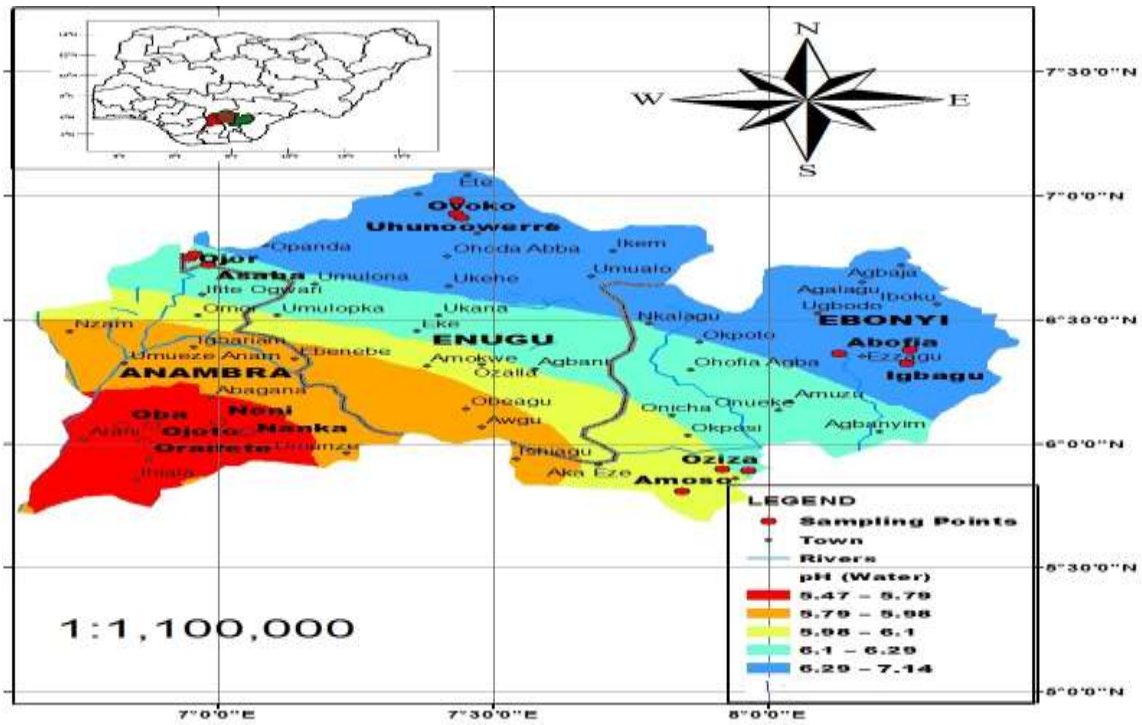


Figure 3: Kriged Map for Percentage Sand Distribution

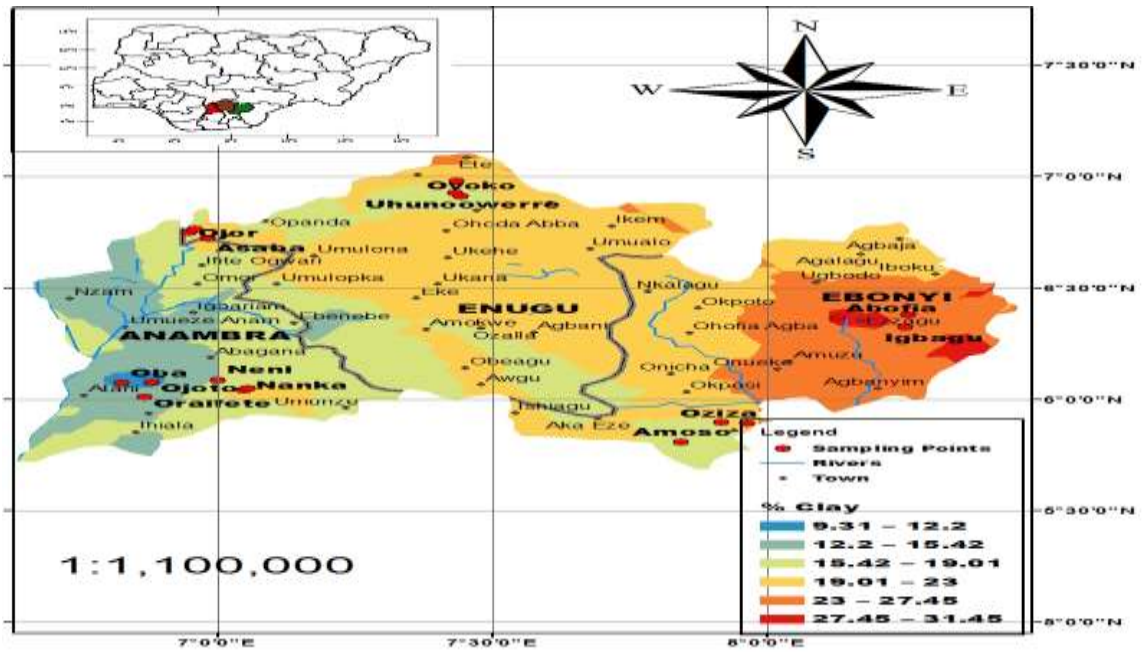


Figure 4: Kriged Map for Percentage Clay Distribution

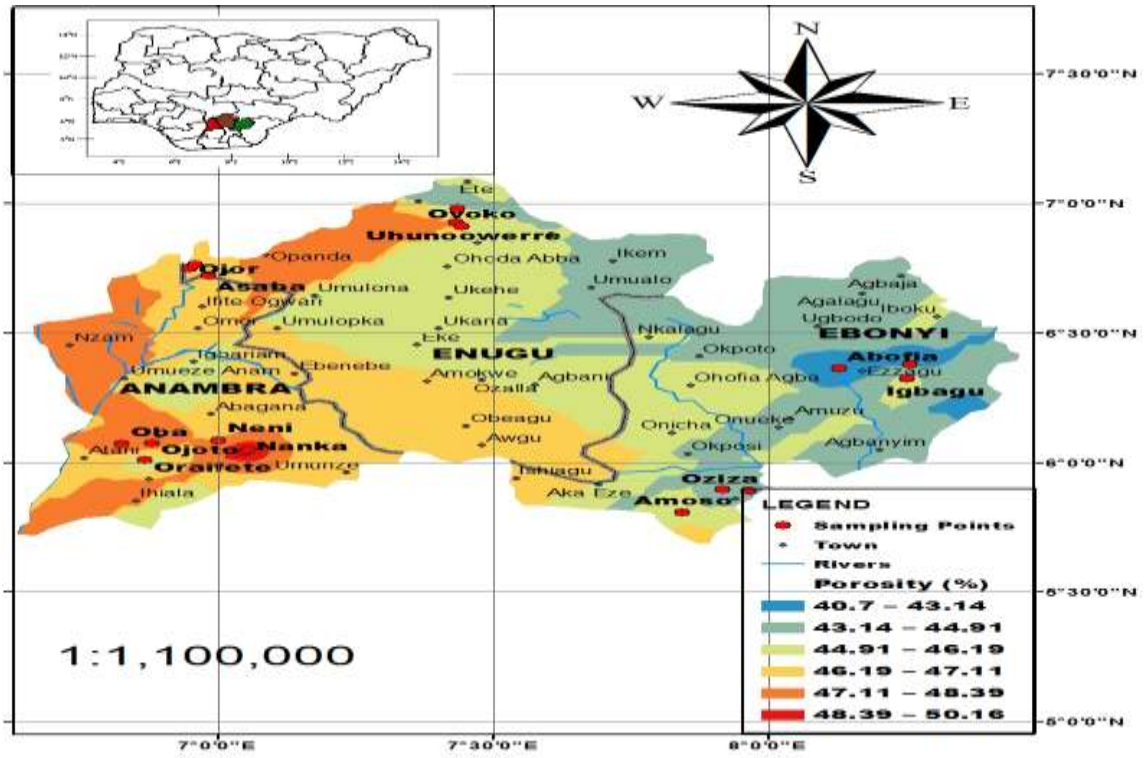


Figure 5: Kriged Map for Porosity

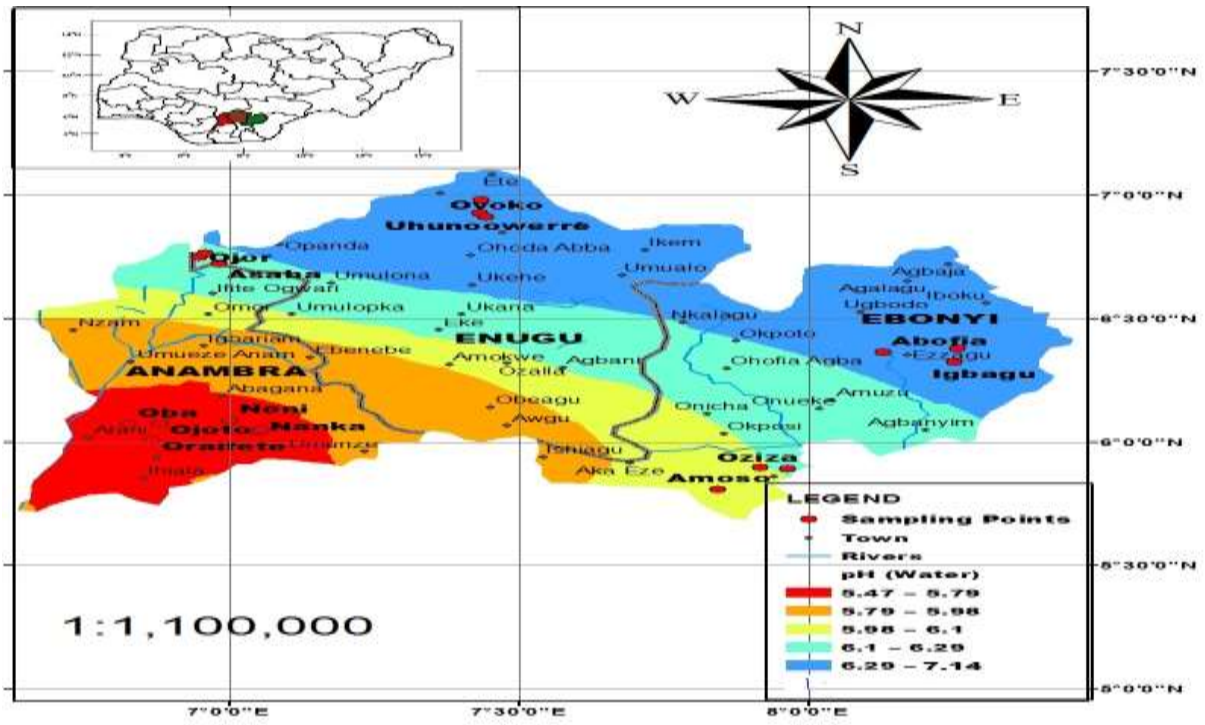


Figure 6: Kriged Map for pH

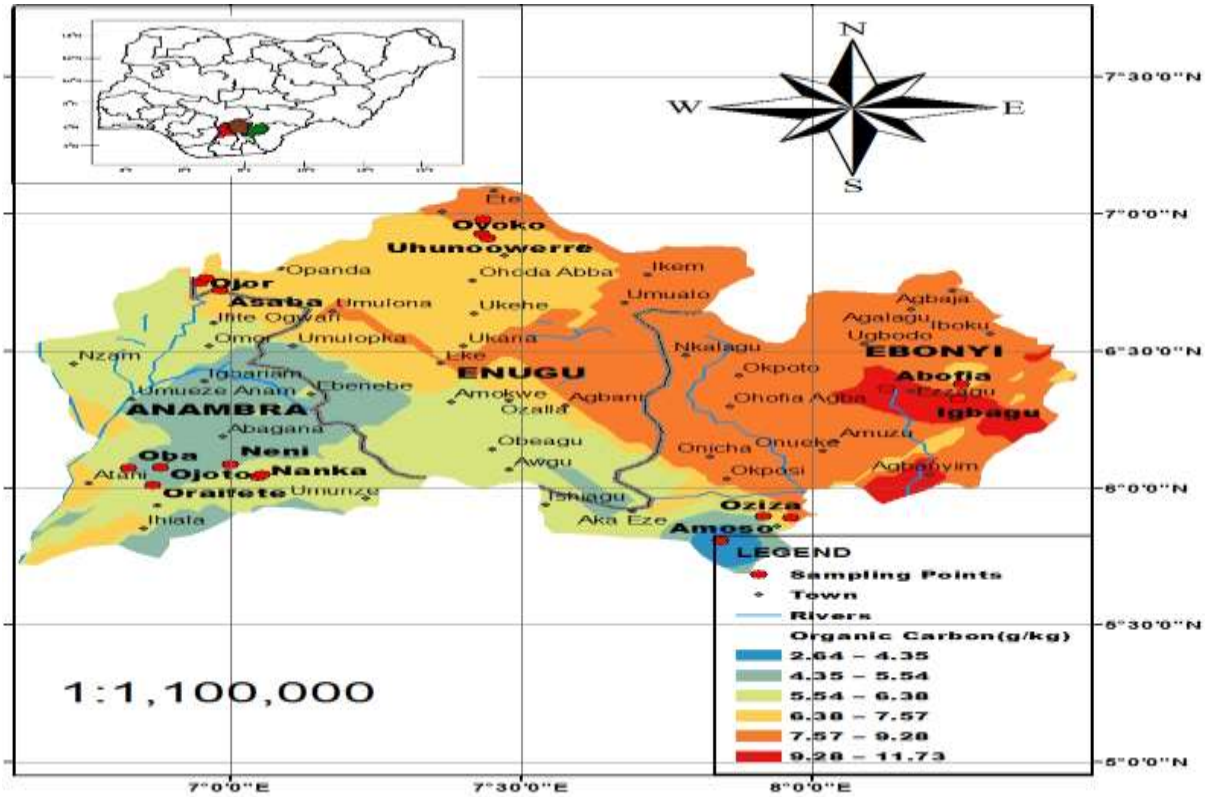


Figure 7: Kriged Map for Organic Carbon

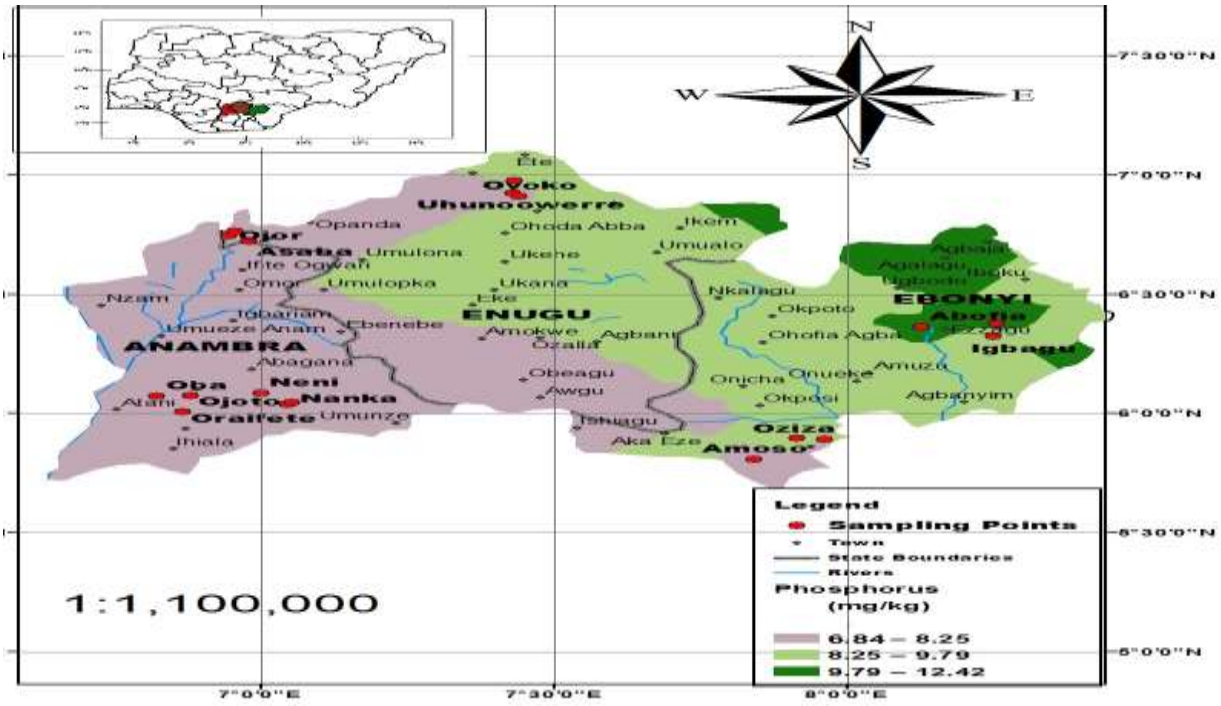


Figure 8: Kriged Map for Phosphorus

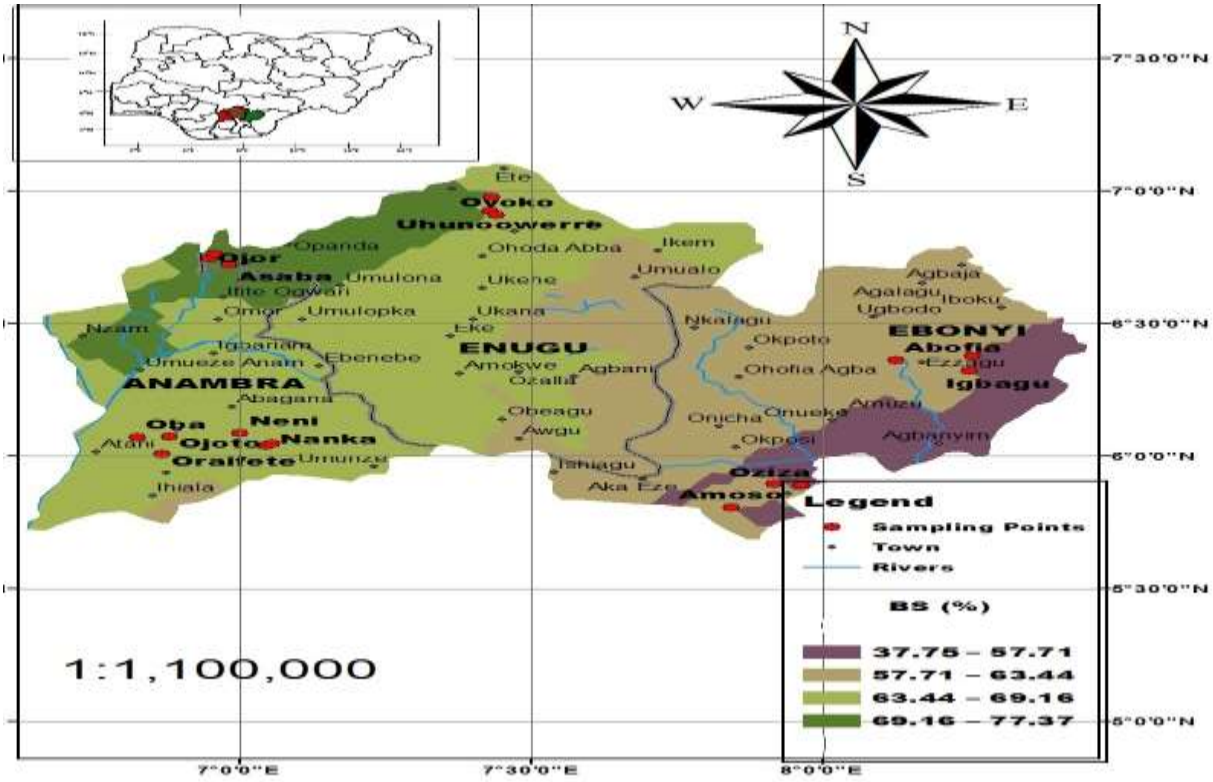


Figure 9: Kriged Map for Base Saturation

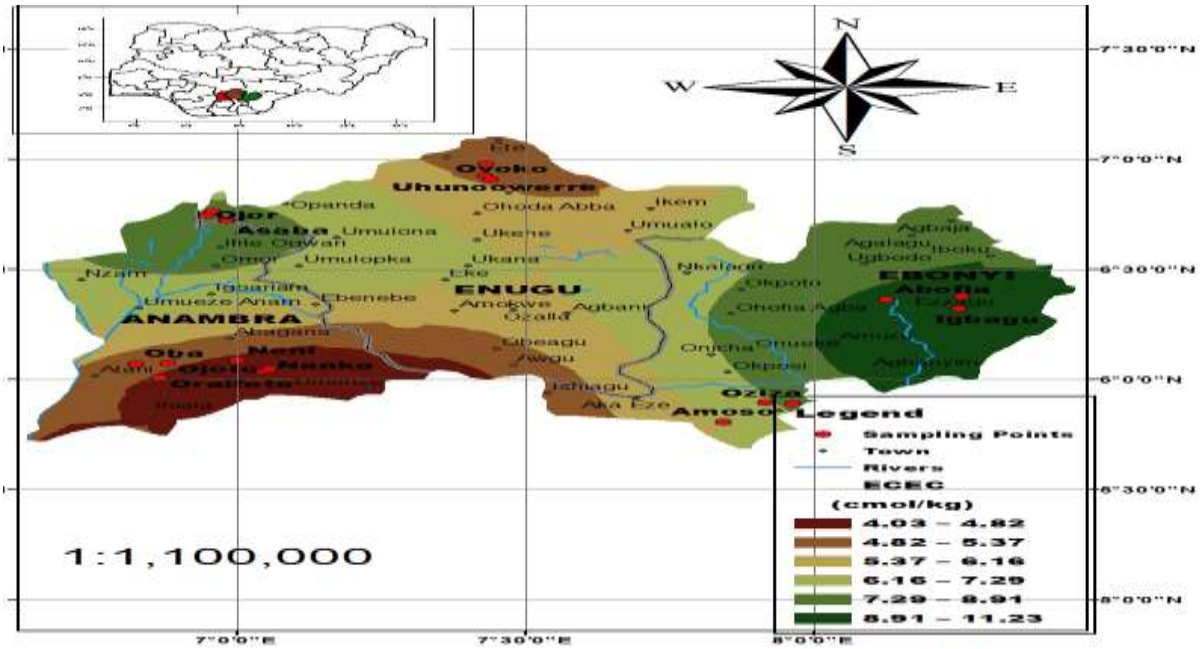


Figure 10: Kriged Map for ECEC

There was also high rate of variation in the base saturation (Figure 10) and is probability caused be structural factors such as parent materials, topography and other natural factors. Higher value of 69.16-77.37% observed in northern part of Anambra and Enugu State. The least value for base saturation was observed in eastern part of Ebonyi State. The value of 63.44-69.16 covered most soils of Enugu State.

The ECEC (Figure 11) followed an observable pattern increasing towards east with the highest value (8.91-11.23 $cmolkg^{-1}$) observed in Ebonyi State. The least value (4.03-4.82 $cmolkg^{-1}$) was observed in the northern part of Anambra State.

Acknowledgement
I sincerely acknowledge the unquantifiable efforts of my supervisors, Prof. E.U. Onweremadu, Dr. (Mrs.) B. N. Aririguzo and Dr (Mrs) C.M. Ahukaemere. I appreciate your support, constructive criticisms and advice.

Conclusion
There is need to understand the spatial distribution of soil properties in a large scale for precision farming and environmental monitoring irrespective of the security challenges facing soil sampling in Southeastern Nigeria. This study showed that few sampling points can be used to predict the value of much large area with some level of accuracy. Geostatistical models were fitted for eight soil properties namely sand content, clay content, porosity, pH, organic carbon, phosphorus base saturation (BS) and effective cation exchangeable capacity (ECEC). Cross validation of semivariogram models through ordinary kriging (OK) method reveals that sand content and ECEC made a good fit and pH and ECEC had the Root mean standardized error (RMSE) less than 1 making good predictions for the unmeasured locations. However, future studies should be made with a standard sampling distance and compare with these results.

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Can Adoption of Nutrition-Sensitive Agriculture Help Narrow Gender Gap on Household Food Security? Evidence from Southwest Nigeria

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Abstract
Household food insecurity has continued to rise on a yearly basis especially in Africa south of Sahara. This increases the risk of adverse health outcome that result from malnutrition if household food insecurity is not addressed. However, nutrition-sensitive agriculture has become a way of addressing nutritional needs in developing countries. This study seeks to investigate if adoption of nutrition-sensitive agriculture could help in narrowing gender gap in household food security using a cross-sectional gender disaggregated survey data collected from 432 farming households across three states in southwest Nigeria. We used household food security as outcome variable which was measure based on five components developed by foreign agencies with little modification to suit our study. These components include measures of food consumption score (FCS), household dietary diversity score (HDDS), coping strategy index (CSI), household food insecurity access scale (HFIAS), and months of adequate household food security (MAHFS). Results show that household size, total farm size, membership of association, access to extension services, awareness of nutrition-sensitive agriculture, and access to training have a positive and statistically significant effect on nutrition-sensitive agriculture adoption among male and female farming households. The outcome variable indicates that the level of food security was higher among the nutrition-sensitive agriculture adopters compared to non-adopters across the gender. Using a robust food security model, we found that there was no statistically significant difference in the level of food security among male and female farming households who had adopted nutrition-sensitive agriculture by all indicators of food security investigated. Therefore, since, adoption of nutrition-sensitive agriculture has improved household food security among the adopters compared to non-adopters. Interventions that are aimed at improving the awareness of nutrition-sensitive agriculture among rural farming households should be encouraged in order close gender gap in households' food security and improve nutrition outcomes.

Keywords: Adoption, food security, farming household heads, gender, nutrition-sensitive agriculture

Introduction
Agriculture can accelerate household access to nutritive foods and contribute to enhancing the diets of all household members. Food insecurity is a central underpinning determinant of nutrition. Individuals having food insecurity has continued to rise on a yearly bases in Africa. According to the FAO report, 426 million of individual experienced moderate food insecurity in 2019 (FAO, ECA, & AUC, 2021). Also, the number of undernourished individuals on the continent has steadily increased throughout the last few years. In recent time, the FAO (2021) reported 250.3 million undernourished individuals in 2019 (FAO et al., 2021). Approximately16.6 million of those live in Northern Africa and 234.7 million in sub-Saharan Africa (FAO et al., 2021). Women and children, and populations that live in areas of conflict, and climate and/ or economic shock are most vulnerable to experience food insecurity. Undernutrition among women and

children remains a major public health problem in utmost low- income and middle- income countries, and is most current in rural areas (Development Initiatives, 2018). Yet, estimates indicate that implementation of 10 nutrition interventions at 90 coverage would only reduce child stunting by 20 (Bhutta et al., 2013), half of the World Health Assembly target of a 40% reduction by 2025. Producing enough food is necessary, but also is guaranteeing equity and access present and for coming generations (Charlton, 2016). Healthy diets were unaffordable for around 3 billion people in the world, particularly the poor, in 2019, given high costs of nutritive food and persistent high degrees of income inequality (FAO and GFRAS, 2021). Still, not only low incomes, but also poor eating habits, lack of knowledge about good nutrition practices, and limited access to different yield add complexity to adequately address this problem (Kachelriess- Matthess et al, 2016).

Across the world, the need for agriculture to champion better nutrition and health has been recognized and was reflected in the dialogues leading up to the United Nations' 2030 Agenda for Sustainable Development (United Nations, 2017), and regionally, it's reflected in the growing number of action to support countries in integrating nutrition interventions into their agriculture investment plans, as illustrated by the Comprehensive Africa Agriculture Development Programme investment plans (Rampa and van Seters, 2013). Countries like Nigeria and Ethiopia, for illustration, have recently developed nutrition-sensitive agriculture plans, a clear manifestation of the major political precedence being given to enhancing the nutritive impact of investments in the agricultural sector. many studies have shown that nutrition-sensitive agriculture interventions can better household food security, nutrition security, undernourishment and have been adopted in various context to address countries' developmental goals (Ruel et al., 2018). According to FAO (2017) defines nutrition sensitive agriculture (NSA) interventions as "a path that seeks to guaranty the production of a variety of affordable, nutritive, culturally appropriate and safe foods in acceptable measure and quality to meet the dietary conditions of populations in a sustainable manner." Many farming interventions developed to advance nutrition outcomes, similar as advancement of biofortified foods, homestead food production and livestock keeping, have bettered household and child dietary intakes (Ruel, 2018). Also, for illustration, the Solar Market Garden program provided solar- power drip irrigation systems in Benin to promote crop diversification to increase access to nutritional foods (Alaofè et al., 2016). Another illustration is the Kutir Rural Management Services Private Limited, which implements value chains and market- led strategies for smallholder farmers to have reliable access to sell their crop (Dubé et al., 2019). Small- scale farmers and households, besides having low incomes, are continually food insecure and suffer habitual or acute forms of malnutrition (Kachelriess- Matthess et al, 2016). Agriculture plays an essential part in guaranteeing nutritive wellbeing, but this should be seen not only for rural populations, but also for the whole society (FAO, 2021). In this way, strategies to enhance livelihoods and income of the poorest sectors of communities are essential to achieve food security for all (Charlton, 2016).

Gender-based inequities within agriculture may limit the sector's capability to deliver nutritive diets and enhance nutrition outcomes (Ruel, 2018). In many low- income rural settings, women have lower control and use of land, livestock and other productive capital (Ruel, 2018), lower economic participation and pay rates (Doss et al., 2015), and heavier workloads (Palacios-López and López, 2015), compared with men. Although it's plausible that closing the gender gap in agriculture could enhance nutritive outcomes, this proposition remains conceptually underdeveloped and reliant on many, generally cited studies (Allendorf, 2007; Duflo and Udry, 2004; Hoddinott and Haddad, 1995) or reviews (Cunningham, 2015; Meinzen- Dick et al., 2019) of women's empowerment, rather than relative measures of gender inequity. Several pathways may exist between gender inequities in agriculture and nutrition outcomes. One pathway may be via women's increased control over household spending decisions, leading to enhanced investment in women's and children's nutrition (Cunningham, 2015). Another pathway may be through advancements in household food availability (Haddad et al., 1996). Studies indicate that agricultural production is more effective in equitable households, leading to enhanced yields overall (Peterman et al., 2010). At the societal status,

gender equity could reduce poverty and enhance nutritional outcomes (Smith et al., 2003) through increases in economic equivalence between households. An ideal gender and nutrition-sensitive agriculture intervention would increase farm productivity, boost women's agency, and promote nutritious crops that can be consumed by women or sold for income without adding workload or infection risk (Kadiyala et al., 2014). Still, there has been no comprehensive documentation on the impact of nutrition-sensitive agriculture which studies a population from a gender household head perspective, similar as the rural household heads in Nigeria, where knowledge and access to nutrition-sensitive agriculture are actually low. There is dearth of knowledge on nutrition-sensitive agriculture especially among rural farming households and by gender at present, thus, this point to a significant gap in the literature that our study aims to fill. therefore, our study fills this vacuum by giving empirical data and provides a first attempt to delve the impact of the nutrition-sensitive agriculture on household food security in Nigeria by gender using quantitative evidence from across- sectional dataset of rural farming households engaging kinds of food crops including home gardening, fruits and vegetables. similar empirical evidence could inform policy makers in sub-Saharan Africa aimed at promoting nutritional crops, eliminate hunger, and boost agricultural productivity. Gender-sensitive agriculture programs, especially those directly targeting women, can increase women's decision-making power around using household resources to buy nutritional foods. Meanwhile, there are immediate policy issues addressed in this study (1) What factors impact rural farming household's decision to engage in nutrition-sensitive agriculture? (2) Is nutrition-sensitive agriculture closing gender gap in household food security? Answering these questions is central to increase households' access to nutritional foods and healthy diets of all household members and upgrade food security in Nigeria and sub-Saharan Africa.

The remainder of the paper is structured as follows. Section 2 discusses applicable literature on nutrition-sensitive agriculture. Section 3 presents the study area, data and sampling techniques, and estimation strategy. Section 4 reports the results and discussion of major findings of the study. Eventually, Section 5 draws conclusion and recommendations from the findings.

Literature Review

While the world had come a long way in enhancing food and nutrition security, the last FAO report (2021) shows a significant lapse in recent times. Global phenomena, such as climate change, political instability and conflict, and economic downturn continue to worsen household food insecurity for vulnerable populations (Boliko, 2019; Wheeler & von Braun, 2013). Further recently, the COVID- 19 pandemic has caused an upset in supply chains leading to an increase in food prices and farming inputs, similar as fertilizer, in low and low-middle income countries, which has resulted in another spike in household food insecurity (World Bank, 2021). A number of reviews of evidence have been published in the past two decades (see, for case, DFID, 2014; Fiorella et al., 2016; Pandey et al., 2016; Webb and Kennedy, 2014), and all of them agree that evidence on what and how agriculture can contribute to nutrition is extremely scarce. The reviews cover a range of agriculture programs including homestead food production systems; home vegetable gardens; biofortified crops; small animals; livestock; fisheries; dairy; and irrigation systems. In spite of differences in the sets of studies reviewed and the approaches and nutrition indicators used in the original studies, the findings from these reviews are amazingly consistent. Also, a systematized literature review on drought and food insecurity in Africa, establish evidence to support that rural populations, especially smallholder farmers, living in areas susceptible to drought remain vulnerable to household food insecurity due to incapability to predict weather for agricultural planning (Ngcamu & Chari, 2020). The unpredictable climate change disrupts indigenous food systems thus reducing food supply for communities. Also, populations living in areas of conflict or political instability are increasingly vulnerable to food security (Anderson et al., 2021). A reason for this is an upset in food production (Anderson et al., 2021; Deaton & Lipka, 2015), which in one of the primary pathways of food security (Deaton & Lipka, 2015). Poverty, gender discrimination and low social status puts women at a greater chance of having food insecurity. In households with food insecurity, mothers have reported giving up their portion of food to

guaranty that children eat. In across-sectional study completed in Ethiopia, being a female head of household was significantly associated with food insecurity (Endale et al., 2014). Food insecurity could also have long term mental health impacts. A qualitative study exploring the experiences of children from food insecure households in Australia establish that their awareness of the household stressors and financial circumstance led to feelings of sadness (Velardo et al., 2021), which can manifest into depression, anxiety or other feeling of distress. In fact, a systematized literature review establishes that there is limited investigation to understand how experiences of food insecurity impacts life course influence on non-communicable disease morbidity and cognitive outcomes (Ward et al., 2018). This may also limit scientific understanding of the health consequences of food insecurity in adolescents into their adulthood. Studies has shown that food insecurity is a gendered experience in which women are at topmost threat for experiencing the negative consequences of food security. This is especially true when the women were the head of household in low and low-middle income countries. A literature review establishes an analysis which stratified the head of household by gender and establish that women head of holds were 75 more likely to experience household food insecurity (Jung et al., 2017). An organized review exploring the impact pathways of nutrition sensitive agricultural and nutritional outcomes establish studies that reported positive effect on food security (Sharma, Di Prima, Essink, & Broerse, 2021). The same regular review reported positive long- term effects on dietary diversity, food consumption and nutrient intake (Sharma et al., 2021). These benefits make them attractive interventions, especially at a global scale because they can be adjusted for the most vulnerable population for food insecurity and malnutrition.

Methodology

Study Area: This study was carried out in Nigeria's southwestern states. These states include Ekiti, Lagos, Ogun, Ondo, Osun, and Oyo States. The entire region is located between latitudes 6° N and 4° S, and longitudes 4° W and 6° E, and covers an area of roughly 114, 271 km^2. The region's typical periodic rainfall is between 1,200 and 1,500 mm, with mean yearly temperatures ranging from 18 to 24°C in the rainy season to 30 to 37°C in the dry season. Due to the area's rich alluvial soil, this part of the country is largely agrarian. Cassava, maize, yam, cocoyam, cowpea, vegetables, and cash crops similar as cocoa, kola nut, rubber, citrus, coffee, cashew, mango, and oil palm are among the notable food crops grown in the area. The study's population included all of the region's rural farming households.

Data and Sampling Methods: In our study, we employed survey data from a cross-section of farming household that was conducted between July to November 2022. The survey involved a multi-stage stratified random sampling method of farming households who grow varieties food crops, vegetables, home garden, and livestock as a primary or secondary occupation. The respondents for the survey were chosen using a four- stage random sampling procedure. The first stage featured a random selection of 50 of the six states in the region, including Ekiti, Ogun, and Oyo. In the second stage, three agricultural zones were randomly picked from each of the three states, for a total of nine agricultural zones. In the third stage, three rural farming communities were chosen at random from each of the nine agricultural zones, for a total of 27 rural farming communities in the study. Eventually, in stage four, 16 rural household heads (8 male and 8 female) were randomly picked from each of the selected rural communities, totalling 432 respondents. Meanwhile, from the sampled households, 6 adopters of nutrition-sensitive agriculture and 10non-adopters of nutrition-sensitive agriculture were eventually selected from each household, totalling 162 adopters and 270non-adopters. The attempt to have equal number of male and female households for the study proved futile, as the study ended up thus, 93 and 69 adopters, 150 and 120non-adopters of nutrition-sensitive agriculture of male and female household heads respectively, were finally interviewed for the study. Our instrument for data collection was structured questionnaire which was deployed on an Android electronic tablet software (Surveybe). The survey instrument was designed based on the objectives of the study. Information was collected from respondents on the socioeconomic characteristics of farm households, various food crop grown, access to information, credit and extension, knowledge on nutrition-sensitive agriculture, and

household food security. Before answering to questions posed by enumerators, respondents' consent was solicited. It was encouraged to all respondents not to engage in the study if they felt uncomfortable, and to withdraw at any time during the survey. Every respondent was duly informed about the study's aim and the significance of partaking in it. We used household food security as outcome variables in this study and it was measure based on five components and these include the household dietary diversity score (HDDS), food consumption score (FCS), coping strategy index (CSI), household food insecurity access scale (HFIAS), and months of adequate household food security (MAHFS, as developed and used by Sahu, Chüzho, and Das, (2017) and modified by Mutea et al. (2019). The treatment variable in the study is the adoption of nutrition-sensitive agriculture by rural farming households measured by a dummy that takes a value of 1 if a rural farming household adopt nutrition-sensitive agriculture, and 0 otherwise. The description of some of the variables included our model are presented in Table 1.

Table 1: Description of variables included in the model

Variables	Description
Outcome variables	
Household dietary diversity score (HDDS)	The number of distinct food categories ingested over 24-hour recall period
Food consumption score (FCS)	The frequency of consumption of 12 different food groups in the last 7 days prior the survey
Coping strategy index (CSI)	The frequency and severity of coping methods used during a food crisis
Household food insecurity access scale (HFIAS)	The frequency-of-occurrence of insufficient food quantity and quality during the last 30 days of recall
Months of adequate household food security)	The number of months food is available or not available in the last 12 months, regardless of the source.
Treatment variables	
Adopt nutrition-sensitive agriculture	Dummy = 1 if household adopted nutrition-sensitive agriculture; 0 otherwise
Control variables	
Age	Age of household head (years)
Marital status	Dummy = 1 if married: 0 otherwise
Education	Years of formal education
Household size	Total number of people in the household
Total farm size	Total farm size cultivated in hectare (ha)
Farming experience	Number of years of farming (years)
Membership of farmers' union	Dummy = 1 if yes; 0 otherwise
Extension service	Dummy = 1 if access to extension service; 0 otherwise
Credit access	Dummy = 1 if access to credit service; 0 otherwise
Awareness of nutrition-sensitive agriculture	Dummy = 1 if aware of nutrition-sensitive agriculture; 0 otherwise
Access to nutrition-sensitive agriculture training	Dummy = 1 if attended any nutrition-sensitive agriculture; 0 otherwise

Source: Field survey (2022)

Estimation strategies

To increase the willingness of farming households to adopt nutrition-sensitive agriculture (NSA) and thus to narrow gender gap in household food security, it is necessary to be aware of the factors that influence the farming household heads' decisions and choices and to understand factors that influence the outcome variable (food security). Following Oyetunde-Usman et al. (2021), suppose the treatment indicator *Ti*

represents the decision to adopt or not adopt nutrition-sensitive agriculture (NSA) by farming household heads which is assumed to be dependent on households' perceived utility. Considering that the adoption of nutrition-sensitive agriculture (NSA) is likely to influence outcome variable (food security) this outcome, however, form a linear function of farming household heads' observed vector of explanatory variables including the dummy variable explaining their choice of adoption which is mathematically expressed as:

$$Y_i = \alpha X_i + \beta T_i + \varepsilon_i \dots\dots\dots\dots\dots\dots (1)$$

where Y_i represents the outcome of household food security; X_i represents the explanatory variable which includes farming household heads' socioeconomic, characteristics (such age, marital status, education, household size, farming experience, income); production characteristics (farm size, labour, cost of input), and institutional characteristics (extension services, access to credit, awareness of nutrition-sensitive agriculture, access to training, membership of association), and the state-wise fixed effects; T_i is the treatment indicator for farming household heads' adoption decisions; α is the vector of parameters of the explanatory variables to be estimated; β represents the vector of parameter estimates of adoption influence of nutrition-sensitive agriculture; and ε_i is the error term.

In this study, the outcome variable is household food security, however the contribution of nutrition-sensitive agriculture (NSA) to the household food security status among male and female farming household heads, was measured using five generally used indicators components household dietary diversity score (HDDS; Swindale and Bilinsky, 2006), food consumption score (FCS; World Food Programme, 2008), coping strategies index (CSI; Maxwell and Caldwell, 2008), and household food insecurity access scale(HFIAS; Coates et al., 2008), and month of adequate household food provisioning (MAHFP; Swindale and Bilinsky, 2010).

Household dietary diversity score (HDDS): is the number of distinct food classes ingested over a specific period, and dietary diversity questions can be answered at both the household and individual levels. Cereals, tubers and roots, vegetables, fruits, meats, eggs, fish and other seafood, legumes, nuts and seeds, milk and milk products, oils and fats, sweets, spices, sauces, and beverages are all divided into 12 food groups. Over the 24-hour recall period, the replies were either consumed "1" or not consumed "0" by the household (FAO, 2012). A household's dietary diversity score ranges from 0 to 12 when all food groups are added together. Values for illustration of different food groups were assigned letters from A to L and classified as either "0" or "1" for the total number of food groups ingested by respondents. The aggregate of food group values was also used to calculate the HDDS for each household head (see equation 2).

$$HDDS = (A + B + C + D + E + F + G + H + I + J + K + L) \dots\dots\dots.. (2)$$

There are no defined cut-off points in terms of the number of food groups that indicate adequate or inadequate dietary diversity for the HDDS, according to Rajendran (2012). As a result, utilizing the equation, the household dietary diversity index (HDDI) was computed to identify low dietary diversity (≤3), medium dietary diversity (4 - 6) and high dietary variety (≥6) (See equation 3).

$$HDD\ Index = \frac{Number\ of\ food\ goups\ consumed\ by\ household\ head}{Total\ number\ of\ food\ groups\ (12)} \times 100\% \dots. (3)$$

Food Consumption Score (FCS): The Food Consumption Score (FCS) for a given household is the frequency- weighted HDDS (IFPRI, 2008), which is used to calculate the frequency of consumption of eight different food types (United States Agency for International Development (USAID), 2015). As a result, 12 different dietary groups were investigated in this study. The Household Food Consumption Score is calculated using a standard 7- day food data set(none = 0, once a week = 1, twice a week = 3, three times

a week = 4, four times a week = 5, and five and more times a week = 6) and by classifying food items into food groups and summing the consumption frequencies of food items within each group(any consumption frequency greater than 7 is recoded as 7, and multiplying the value obtained for each food group by its weight). The nutritional density of the food groups serves as the guiding element for determining weight. Cereals = 2, Vegetables = 1, Fruits = 1, Meat and fish = 4, Pulses = 3, Milk = 4, Oils = 0.5, Sugar = 0.5, and Condiments = 0. adding all food groups provides a household food consumption score. therefore, a typical formula for calculating FCS is shown in equation (4).

$$FCS = (cereals * 2) + (legumes * 3) + vegetables + fruits + (flesh\ \&\ organ\ meat * 4) + (roots\ \&\ tubers) + (eggs) + (fish\ \&\ seed\ foods) + (milk\ \&\ dairy\ products * 4) + (fats * 0.5) + (sugar * 0.5) + (spices, condiments\ \&\ beverages) \ldots\ldots\ldots\ldots\ldots\ldots (4)$$

Then finally the food consumption score were categorized into: 0 - 46 (poor), 46.5 - 92 (borderline), and above 92 (acceptable) (FAO, 2011).

Coping Strategy Index (CSI): The World Food Programme created this indicator, which is a weighted score that looks at the frequency and severity of coping approaches during a food crisis. CSI could be determined using various weekly food possessions per household member for food types consumed over some time, i.e., the frequency of each coping strategy response multiplied by its weight. Weights are calculated based on qualitative observations or focus group discussions, similar as when buying food on credit. A household coping strategy index is created by adding all of the responses together. The coping strategy index was thus grouped into 0 – 2 = No or low coping (Food secure), 3 – 12 = Mildly food insecure, and ≥ 13 = High coping (Moderately/ severely food insecure).

Household Food Insecure Access Scale (HFIAS): This tool assesses insufficient food quality and quantity, as well as anxiety over food or market security. The HFIAS measures a combination of psychological and adequacy criteria. The test was created and is used by the United States Agency for International Development (USAID) to assess food security. It includes nine poor food status occurrence questions with yes/ no answers and another set of nine questions on frequency- of- occurrence during the last 30 days of recall (Coates, Swindale, and Bilinsky, 2007). Some of the nine questions, for illustration, were employed in the development of the household food insecurity access scale (HFIAS) model, and these questions were administered using a dummy method, with respondents anticipated to answer yes or no. The responses were used to construct raw food security ratings ranging from 0 to 9, with 0 indicating the most food-secure families and 9 indicating the most food-insecure households in this study (Melgar-Quinonez, et al., 2006, and Sseguya, 2009). The scale was also classified into four food security classes (1 = food secure, 2 = mildly food insecure, 3 = moderate food insecure, and 4 = severely food insecure) based on each household response.

Months of Adequate Household Food Provisioning (MAHFP): This tool determines whether or not there was restricted food availability in the previous 12 months, regardless of the source. This can be calculated as 12 months minus the total number of months in the previous 12 months when a household was unfit to meet its food demands. We estimated the mean for all the homes in the sample and classified those who were above the mean as food secure and those who were below the mean as food insecure (i.e., 10 – 12 months = food secure and 0 – 9 months = food insecure).

Computation of Food Security Index

To attain food security, a household (whether male or female-headed) must meet all food security dimensions at the same time. To meet algebraic requirements, we first standardized each of the original indicator scores into a Z-score for each household (see Equation 6), then aggregated the indicators and used

correlation analysis to determine the degree of linearity between the indicators of the food security index (FSI) and the final composite FSI. The standardization method was used to normalize all indicators in the food security index using the non-weighted approach to make all indicator values consistent and congruent. Depending on the type of data, the indicators were standardized to fit within the range of zero (0) to one (1) using either linear normalization or Z-score. The normalized scores or Z-scores for each indicator were calculated in this study using equation (5):

$$z_n = \frac{x_n - \mu_n}{\sigma_n} \dots\dots\dots\dots\dots\dots\dots (5)$$

where z_n = *normalized score or Z-score for each food security indicator for the n[th] household*

x_n = *original score of an indicator*

μ_n = *mean of the original distribution*

σ_n = *standard deviation of the original distribution of household n*

We used Sahu et al. (2017) to aggregate the standardized values of all the indicators, with a minor change that employs the difference between a minimum of food security (HDDS, FCS, and MAHFP) and a maximum of food insecurity (CSI and HFIAS) indicators to generate a composite food security index (FSI). Since food security increases with household dietary diversity, food consumption and months of adequate household food provision but reduces with coping strategies and household food insecure access scale, therefore the Composited Food Security Index (x_n^{FSI}) of a household in each state is expressed as:

$$x_n^{FSI} = [(z_n^{HDDS} + z_n^{FCS} + z_n^{MAHFP} + (-z_n^{CSI}) + (-z_n^{HFIAS})] \dots\dots (6)$$

Where, x_n^{FSI}= composite food security index for an *n*th farming household in a state *i*;

z_n^{HDDS}= normalized value of HDDS;

z_n^{FCS}= normalized value of FCS;

z_n^{MAHFP}= normalized value of MAHFP;

z_n^{CSI} = normalized value of CSI and

z_n^{HFIAS}= normalized value of HFIAS.

The composite food security index, on the other hand, ranges from 0 to 1, with 1 indicating the highest level of food security and 0 signifying no food security at all.

A basic ranking of the households based on the indices viz., $\overline{y_i}$ will enough for categorization. Furthermore, proper fractile categorization from an assumed probability distribution is required for a meaningful definition of the various levels of food security. The Beta distribution, which takes values in the interval (0, 1) and is given by, is a probability distribution that is acceptable for our topic.

$$f(z) = \frac{z^{a-1}(1-z)^{b-1}}{\beta(a,b)}, 0 < z < 1 \; and \; a, b > 0 \dots\dots\dots (7)$$

Where $\beta(a,b)$ is the beta function defined by

$$\beta(a,b) = \int_0^1 x^{a-1}(1-x)^{b-1} \dots\dots\dots\dots\dots\dots (8)$$

The Beta distribution is skewed. Assuming$(0, z_1)$ and $(z_1 z_2)$ be the linear intervals such that each interval has the same probability weight of 50 per cent. Therefore, the fractile intervals can be used to categorize the food security into; food secure (if $0<\overline{y_i} < z_1$), and food insecure (if $z_1<\overline{y_i} < z_2$).

Results and Discussion

Summary of descriptive statistics

Table 2 present descriptive statistics of covariates that we assumed to influence the adoption of nutrition-sensitive agriculture among the male and female farming household in the study area. These covariates

include age, marital status, education, household size, total farm size, farming experience, membership of association, access to extension, access to credit, awareness of nutrition-sensitive agriculture, and access to nutrition training. The results show statistically significant differences in most of the covariates between nutrition-sensitive agriculture adopters and non-adopters of male and female farming household heads' characteristics. The nutrition-sensitive agriculture adopters are relatively older, married with basic education, larger household and farm size with more farming experience among both male and female farming households. But they relatively have less access to institutions such as extension services, credit facilities, awareness, and nutrition-sensitive agriculture training programs compared with non-adopters of nutrition-sensitive agriculture. For instance, about 88% of the nutrition-sensitive agriculture adopters are members of farmers association compared with 67% of non-adopters among male farming households. In female category, 83% of adopters are members of farmers' union while only 52% of non-adopters belongs to farmers associations. Also, 46% of adopters of nutrition-sensitive agriculture among male farming households have more access to extension services compared with about 36% of non-adopters. Likewise, among female farming households only 39% of nutrition-sensitive agriculture adopters have access to extension services compared with just 25% of non-adopters. Results show that only 43% and 34% of nutrition-sensitive agriculture adopters in both male and female farming households have access to credit facilities compared with about 31% and 22% of non-adopters respectively. About 100% and nearly 89% of adopters are aware of various nutrition-sensitive agriculture practices compared with 37% and 26% of non-adopters among male and female farming households. Moreover, majority (73%) of nutrition-sensitive agriculture adopters in male farming households received training on nutrition-sensitive agriculture practices compared with just 69% of the adopters among female farming households. However, our findings suggest that adopters and non-adopters of nutrition-sensitive agriculture among male and female farming households are significantly different.

Table 2. Summary descriptive statistics by treatment

Variables	Male farming households					Female farming households				
	Full sample (N=243)	Adopters (n=93)	Non-adopters (n=150)	Mean difference (2-3)	t-Test (p-value)	Full sample (N=189)	Adopters (n=69)	Non-adopters (n=120)	Mean difference (3-4)	t-Test (p-value)
		1	2				3	4		
Age	54.61 (15.018)	56.24 (14.319)	52.96 (11.278)	3.28	2.368 (0.011)***	53.21 (13.215)	51.89 (10.327)	54.65 (12.315)	-2.76	2.765 (0.000)***
Marital status	0.81 (0.365)	0.85 (0.357)	0.76 (0.406)	0.09	0.649 (0.366)	0.72 (0.259)	0.69 (0.421)	0.74 (0.316)	-0.05	ne1.923 (0.000)***
Education	7.65 (5.245)	8.59 (4.395)	6.69 (0.495)	1.90	2.052 (0.002)***	6.56 (4.929)	6.87 (4.514)	6.24 (4.646)	0.63	1.618 (0.107)*
Household size	10.22 (4.642)	11.19 (4.836)	9.25 (3.423)	1.94	1.791 (0.000)***	8.81 (3.191)	8.15 (2.309)	9.47 (1.369)	-1.47	2.124 (0.000)***
Total farm size	7.22 (4.331)	9.05 (2.796)	5.36 (2.551)	3.69	4.143 (0.000)***	3.94 (1.163)	4.26 (1.217)	3.62 (0.388)	0.64	2.056 (0.000)***
Farming experience	32.31 (14.679)	35.21 (12.145)	29.38 (10.601)	5.83	6.284 (0.001)***	28.84 (5.036)	30.49 (6.594)	27.19 (6.439)	3.30	4.182 (0.030)***
Membership of farmers' union	0.78 (0.467)	0.88 (0.282)	0.67 (0.630)	0.21	2.817 (0.000)***	0.68 (0.455)	0.83 (0.397)	0.52 (0.503)	0.31	2.960 (0.000)***
Extension service	0.41 (0.493)	0.46 (0.442)	0.36 (0.417)	0.10	0.257 (0.000)***	0.32 (0.472)	0.39 (0.439)	0.25 (0.527)	0.14	0.451 (0.080)***
Credit access	0.37 (0.455)	0.43 (0.294)	0.31 (0.521)	0.12	1.076 (0.000)***	0.27 (0.373)	0.34 (0.662)	0.22 (0.336)	0.12	2.157 (0.032)***
Awareness of nutrition-sensitive agriculture	0.67 (0.690)	1.00 (0.000)	0.37 (0.329)	0.63	0.135 (0.000)***	0.58 (0.439)	0.89 (0.092)	0.26 (0.456)	0.63	0.192 (0.000)***
Access to NSA training	0.48 (0.519)	0.73 (0.398)	0.24 (0.406)	0.49	0.347 (0.000)***	0.43 (0.526)	0.69 (0.572)	0.18 (0.137)	0.51	0.513 (0.000)***

The t-test was carried out to test for difference in covariates between of adopters and non-adopters of nutrition-sensitive agriculture among male and female farming households;

*, **, ***represent statistically significant at 10, 5 and 1%, respectively.

Source: Field survey (2022)

Determinants of adoption of nutrition-sensitive agriculture among male and female farming households

In this study, we employed Probit model to investigate the major determinants of adoption of nutrition-sensitive agriculture among male and female farming households as shown in Table 3. The coefficient and marginal effect from the maximum likelihood estimates of the probit model were used in explaining the effect of the covariates included in the model on decision of male and female farming households to adopt or not the nutrition-sensitive agriculture. The marginal effect, on the other hand, is more effective than the coefficient to characterize the magnitude of a probabilistic model. This is because the sign and magnitude of the marginal effect determine the direction and magnitude of the likelihood influence of covariates on their decision to adopt or not (Lokshin, 2004). The marginal effect is thus, used in explaining the results of the model. The Pseudo-R^2, statistical significance of the log-likelihood (LR) ratio, and chi-square probability in Table 3 indicate that the specified model is compatible and best fit for the model used in the study. Results in Table 3 show that six of the eleven covariates included in the model are statistically significant in explaining the decision of male farming household heads to adopt nutrition-sensitive agriculture. These include household size, total farm size, membership of association, access to extension services, awareness of nutrition-sensitive agriculture, and access to training. Meanwhile among female farming households sampled only five of the eleven covariates are statistically significant in explaining the decision of household to adopt or not. Some of these covariates are household size, total farm size, membership of association, awareness of nutrition-sensitive agriculture, and access to training.

Table 3: Determinants of adoption of nutrition-sensitive agriculture

Variable	Male farming households				Female farming households			
	Probit regression		Marginal effects		Probit regression		Marginal effects	
	Coefficient	Std. error	dy/dx	Std. error	Coefficient	Std. error	dy/dx	Std. error
Age	-0.026	0.016	-0.005	0.003	0.014	0.019	0.008	0.006
Marital status	-0.523	0.491	-0.277	0.216	-1.457	0.575	-0.512	0.214
Education	0.061	0.027	0.005	0.012	0.018	0.033	-0.002	0.010
Household size	0.157*	0.033	0.031*	0.011	0.293**	0.091	0.074***	0.032
Total farm size	0.936***	0.116	0.452***	0.041	1.145*	0.279	0.247***	0.104
Farming experience	0.027	0.018	0.027	0.005	0.122	0.014	0.004	0.006
Membership of farmers' union	1.854***	0.513	0.306***	0.047	1.041***	0.226	0.289***	0.093
Extension service	0.572**	0.272	0.175**	0.091	0.063	0.125	0.017	0.098
Credit access	0.558	0.134	0.178	0.126	0.392	0.337	0.025	0.120
Awareness of nutrition-sensitive agriculture	0.968***	0.305	0.439***	0.075	2.713***	0.212	0.395***	0.065
Access to NSA training	1.471***	0.394	0.268***	0.083	1.246***	0.321	0.321***	0.123
_constant	-5.198	1.167			-7.253	1.469		

Male: Log likelihood = -48.10867; Pseudo R^2 = 0.8262; LR chi2 (11) = 387.69; Prob>chi^2 = 0.000; Observation = 243

Female: Log likelihood = -45.63842; Pseudo R^2 = 0.6884; LR chi2 (12) = 259.75; Prob>chi^2 = 0.000; Observation = 189

*, **, ***represent statistically significant at 10, 5 and 1%, respectively.

Source: Author's computation (2022).

The coefficient of household size is positive and significantly associated with the adoption of nutrition-sensitive agriculture among both male and female farming households at 10 and 5%, respectively. This implies that an increase in the number of household members will influence adoption decision of the household heads. Also, the total farm size coefficient is positively and statistically significant and explains the likelihood of nutrition-sensitive agriculture adoption among both categories farming households. The marginal effect of a unit increase in total farm size on the conditional probability of nutrition-sensitive agriculture adoption are 0.452 and 0.247 for male and female farming households, which suggests that an additional increase in total farm size will lead to a 45.2 and 24.7 percent increase in nutrition-sensitive agriculture adoption among both categories of farming households. A possible explanation is that most farming households either male or female headed with large farm size are more like to adopt nutrition-sensitive agriculture than those with small farm size and which may be due to other institutional supports getting from external sources including government. Similar to our findings, size of cultivated land was found to positively affect agricultural technology use in Ghana (Nata et al., 2014), and Makate et al. (2019) opined that size of land owned by a farmer is also found to have a positive effect on adoption of multiple Climate Smart Agricultural (CSA) innovations in Southern Africa. Furthermore, membership of farmers association increases farming households' likelihood of adopting nutrition-sensitive agriculture. The coefficient of membership of association is positive and significantly influenced the likelihood of both male and female farming households' decision to adopt nutrition-sensitive agriculture. The marginal effect shows that being a member of association increases the probability of adopting nutrition-sensitive agriculture by 30.6 and 28.9 percent respectively, in male and female farming households. One plausible explanation for this could be improved access to information and knowledge sharing among members in the social groups. These findings are in line with previous studies (Daadi and Latacz-Lohmann, 2021). Results in Table 3 indicated a significant positive influence of access to extension on the adoption of nutrition-sensitive agriculture among male farming households. The results suggest that households who had access to extension services are 17.5% likely to adopt nutrition-sensitive agriculture. Also, level of awareness had a positive and significant relationship with the decision of male and female farming households to adopt nutrition-sensitive agriculture. This indicates that an increase in enlightenment campaign and sensitization is likely to increase adoption level. The marginal effects imply that there is a probability of adoption to increase by 43.9% and 39.5% if awareness is raised among male and female farming households respectively. This is in line with a priori expectations indicating that increased awareness is a key component driving the technology adoption decision process. Access to training was found positive and significant associated with adoption of nutrition-sensitive agriculture among male and female farming households. This suggests that increased access to capacity building training will improve farming households' knowledge on nutrition-sensitive agriculture thereby, increase adoption, health benefits and household food security.

Influence of adoption of nutrition-sensitive agriculture to male and female farming households' food security

The influence of adoption of nutrition-sensitive agriculture to food security among male and female farming households was estimated using five most widely used indicators/components of food security and these include; household dietary diversity score (HDDS), food consumption score (FCS), coping strategies index (CSI), household food insecurity access scale (HFIAS), and months of adequate household food provisioning (MAHFP).

Food consumption score (FCS) or pattern of male and female farming households

Table 4 show that 5.4% and 11.6% of male and female farming households who have adopted nutrition-sensitive agriculture had poor food consumption scores compared to their non-adopters' counterparts with 24.7% and 24.2% food consumption score, respectively. Also, the household food consumption score indicates that 31.2 and 27.5% of the nutrition-sensitive agriculture adopters among male and female farming

households were at the borderline while more than half (54.0 and 55.0%) of non-adopters fell within this category (borderline). Results show that majority (63.4 and 60.9%) of male and female farming households of nutrition-sensitive agriculture adopters had adequate dietary level of food consumption score whilst only 21.3 and 20.8% of non-adopters were also at this level. This suggests that most adopters of nutrition-sensitive agriculture among male and female farming households had an acceptable food consumption score compared to their non-adopters. One possible explanation for this is that the practice of nutrition-sensitive agriculture has positive impact on the household food consumption or pattern of the adopters.

Table 4: Classification of male and female farming households by their food consumption score (FCS) over 7 days prior to survey

Categorization	HFC Index	Male farming households				Female farming households			
		Adopters		Non-adopters		Adopters		Non-adopters	
		(f)	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*
Poor	0 – 46	5	5.4	37	24.7	8	11.6	29	24.2
Borderline	46.5 – 92	29	31.2	81	54.0	19	27.5	66	55.0
Acceptable	≥92	59	63.4	32	21.3	42	60.9	25	20.8
Total		**93**	**100**	**150**	**100**	**69**	**100**	**120**	**100**

Source: Field survey (2022); f= frequency; %= percentage

Household Dietary Diversity Score (HDDS)

Data on household dietary diversity was collected based on several food groups consumed throughout 24-hour recall prior the survey. Results in Table 5 show that among the nutrition-sensitive agriculture adopters, only (7.5 and 8.9%) of male and female farming households had consumed up to 3 food groups in the last 24 hours before this survey and they are regarded as low dietary diversity (LDD) compared to 33.3 and 20% of non-adopters. About 34.4% of male and 29% of female among the adopters had consumed between 4 to 6 food groups while most (44.7% and 54.2%) of their non-adopters' counterparts fell into this category and were considered as medium dietary diversity (MDD). However, more than half (58.1 and 62.3%) of male and female farming households in adopter's category had high dietary diversity that is, they had consumed more than 6 food groups in the last 24 hours as compared to only 20% and 25.8% of their non-adopters in the study area. The influence of adoption of nutrition-sensitive agriculture also, reflected on dietary diversity score among adopters in both categories of farming households.

Table 5: Distribution of farming household by dietary diversity score

Categorization	HDD Index (HDDI)	Male farming households				Female farming households			
		Adopters		Non-adopters		Adopters		Non-adopters	
		(f)	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*
Low Dietary Diversity (LDD)	≤3 food groups	7	7.5	50	33.3	6	8.9	24	20.0
Medium Dietary Diversity (MDD)	4 - 6 food groups	32	34.4	67	44.7	20	29.0	65	54.2
High Dietary Diversity (HDD)	≥6 food groups	54	58.1	33	22.0	43	62.3	31	25.8
Total		**93**	**100**	**150**	**100**	**69**	**100**	**120**	**100**

Source: Field survey (2022); f= frequency; %= percentage

Coping Strategies used by male and female respondents

The results in Table 6 present various coping strategies adopted among male and female farming households during the period of food insecurity. Table 2 show that 67.4 % and 53.5% of male and female farming households under nutrition-sensitive agriculture's adoption category have relied on less preferred and less expensive foods compared to their non-adopters (52.3 and 60.2%) counterparts. Also, among the adopters in both categories of farming households 45.7% and 66.9% of male and female respectively bought food on credit, 40.8% of male and 57.1% of female reduced the quantity of meals taken per day, while more than half (63.3% male and 54.6% female) reduces the number of times they eat in their house. Also, 25.9% of male and 46.5% of female farming households resulted into sales of their assets like land, 59.5% of male and 38.4% of female borrowed money from their friends and relatives, while most (73.1% and 60.7%) of male and female respectively, cut down expenditure on food items. Some 64.6% of male and 52.3% of female farming households engaged in pre-mature harvesting of food crops from farm while 55.6% of male and 68.2% of female engaged in off-farm activities.

Table 6: Distribution of farming households by coping strategies adopted

Coping strategies used among farming households	Male farming households		Female farming households	
	Adopters (%)	Non-adopters (%)	Adopters (%)	Non-adopters (%)
Reliance on less preferred and less expensive foods	67.4	52.3	53.5	60.2
Buying food on credit	45.7	41.1	66.9	74.5
Reduce the quantity of meals taken	40.8	52.5	57.1	69.3
Reduce the number of times to eat	63.3	70.6	54.6	62.1
Sales of assets such as land, jewelleries	25.9	49.3	46.5	57.8
Borrow money from friends/relatives	59.5	66.3	38.4	42.2
Cut down expenditure on food items	73.1	78.9	60.7	65.5
Pre-mature harvesting of food crops from farm	64.6	71.5	52.3	60.9
Engaging in off-farm activities	55.8	62.6	68.2	84.7

Source: Field survey (2022); %= percentage

Categorization of farming households based on coping strategies adopted during the period of food insecurity

Table 7 show that among the adopters of nutrition-sensitive agriculture, 15.1 and 117.4% of male and female farming households employed a high number of coping strategies compared to 30.7% and 30.8% of their non-adopters' counterparts during a period of food insecurity and are classified as food insecure, while 29.0% and 34.8% of adopters among male and female farming households respectively employed coping mechanism ranging from 3 to 12 as against 46% of male and 39.2% of female in non-adopters category and thus, could be regarded as mildly food secure. Also, results in Table 7 indicate that most (55.9% and 47.8%) of male and female farming household who had adopted nutrition-sensitive agriculture employed at most two coping strategies and are therefore classified as food secure. These findings suggest that adoption of nutrition-sensitive agriculture practices has helped the adopters in male and female farming households to reduced their coping strategies during the period of food scarcity.

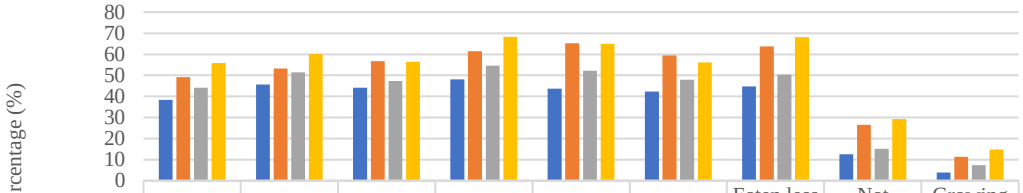
Table 7: Distribution of farming households coping strategy index

Categorization	Coping Strategy Index (CSI)	Male farming households				Female farming households			
		Adopters		Non-adopters		Adopters		Non-adopters	
		(f)	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*
Low or No coping (food secure)	0 – 2	52	55.9	35	23.3	33	47.8	36	30.0
Mildly food secure	3 – 12	27	29.0	69	46.0	24	34.8	47	39.2
High coping (food insecure)	≥13	14	15.1	46	30.7	12	17.4	37	30.8
Total		**93**	**100**	**150**	**100**	**69**	**100**	**120**	**100**

Source: Field survey (2022); f= frequency; %= percentage

Household Food Insecurity Access Scale (HFIAS) of respondents

Figure 1 presents household food insecurity access scale questions posed to respondents in the study area. The results indicate that among the nutrition-sensitive agriculture adopters, 38.4 and 44.1% of male and female farming households respectively worried about getting enough food to eat during the period of food insecurity compared to 49.2 and 55.8% among the non-adopters' counterparts. Some adopters (45.6 and 51.5%) of male and female farming households actually failed to get enough food when compared with 53.3 and 60.1% of their non-adopters, while 44.2% of male and 47.4% of female under adopter's category ate poor quality foods during the period of food insecurity. Also, 48.1 and 54.6% of male and female respondents among the nutrition-sensitive agriculture adopters relied on a few kinds of foods, some (43.7 and 52.2%) of male and female adopters reduced the amount of food eaten with most (65.2 and 64.9%) of their male and female non-adopters fell into this situation. Findings reveal that about 42.3% of male and 47.9% of female farming households among the adopters are skipping meals during the period of food insecurity. More results show that some 44.7% of male and half (50.3%) of female adopters eating less than what one feels they should have eaten per day, 12.5% of male and 15.1% of female adopters did not eat for a day because of lack of food compared to 26.5 and 29.3% of their non-adopters' counterparts, while only 3.9 and 7.4% of them growing thinner because of not eating enough food during the period of food insecurity.



	Worrying about getting enough food	Actual failure to get enough food	Eating poor quality foods	Relying on a few kinds of foods	Reducing the amount of food eaten	Skipping meals	Eaten less than what one feels they should have eaten	Not eating for a whole day because of lack of food	Growing thinner because of not eating enough food
Male Adopters (%)	38.4	45.6	44.2	48.1	43.7	42.3	44.7	12.5	3.9
Male Non-adopters (%)	49.2	53.3	56.7	61.4	65.2	59.5	63.8	26.5	11.3
Female Adopters (%)	44.1	51.5	47.4	54.6	52.2	47.9	50.3	15.1	7.4
Female Non-adopters (%)	55.8	60.1	56.5	68.3	64.9	56.2	68.1	29.3	14.8

Fig. 1: Distribution of farming households by food insecurity access scale
Source: Field survey (2022)

Classification of farming households based on household food insecurity access scale (HFIAS) during the period of food scarcity

Results in Table 8 show that among the adopters of nutrition-sensitive agriculture only 5.4% and 4.3% of male and female respondents respectively are classified as severely food insecure compared to 20% of male and 15% of female non-adopters who fall under this category. Results also show that 9.7% of male and 10.2% of female among the adopters were moderately food insecure while some (29.0%) of male and 30.0% of female respondents who had adopted nutrition-sensitive agriculture were classified into mildly food insecure in the study area. Findings show that more than half (55.9 and 53.6%) of male and female adopters are categorized as food secure whilst only 17.3% and 19.2% of male and female non-adopters fell into this category. The implication of having half of the adopters among male and female farming households in the food-secure category shows some substantial amount of impact of adoption of nutrition-sensitive agriculture on their household food security.

Table 8: Distribution of farming households based on food insecurity access scale

Categorization	Household Food Insecurity Access Scale Index (HFIASI)	Male farming households				Female farming households			
		Adopters		Non-adopters		Adopters		Non-adopters	
		(f)	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*
Food secure	1	52	55.9	26	17.3	37	53.6	23	19.2
Mildly food insecure	2	27	29.0	43	28.7	22	31.9	36	30.0
Moderately food insecure	3	9	9.7	51	34.0	7	10.2	43	35.8
Severely food insecure	4	5	5.4	30	20.0	3	4.3	18	15.0
Total		**93**	**100**	**150**	**100**	**69**	**100**	**120**	**100**

Source: Field survey (2022); f= frequency; %= percentage

Months of Adequate Household Food Provisioning (MAHFP)

Table 9 show that most (48.4 and 46.4%) of the male and female farming households among the adopters of nutrition-sensitive agriculture had between 10 and 12 months of adequate provisioning indicating a very high food security compared to few (20.7% of male and 19.2% of female) of their non-adopters' counterparts who fell into this category. Also, results indicate that 36.6 and 36.3% of the male and female farming households respectively among the adopters are highly food secure with adequate food for 7–9 months, some 11.8 and 11.6% of them were moderately food insecure with adequate food for 4–6 months while only 3.2% of the male and 5.7% of the female farming households were severely food insecure with adequate food less than 3 months a year. Our findings suggest that adopters of nutrition-sensitive agriculture had adequate access to food based on number of months per year more than non-adopters among the male and female farming households thus, implying influence of adoption.

Table 9: Distribution of farming households by months of adequate food provisioning

Categorization	Month of Adequate Household Food Provisioning Index (MAHFPI)	Male farming households				Female farming households			
		Adopters		Non-adopters		Adopters		Non-adopters	
		(f)	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*
Very high	10 – 12	45	48.4	31	20.7	32	46.4	23	19.2
High	7 – 9	34	36.6	37	24.7	25	36.3	29	24.2
Medium	4 – 6	11	11.8	49	32.6	8	11.6	37	30.8
Low	0 – 3	3	3.2	33	22.0	4	5.7	31	25.8
Total		**93**	**100**	**150**	**100**	**69**	**100**	**120**	**100**

Source: Field survey (2022); f= frequency; %= percentage

Food Security Index by farming households

Results in Table 10 present the composite food security index of the male and female farming households based on the adoption of nutrition-sensitive agriculture in the study area. Results show that male and female farming households who had adopted nutrition-sensitive agriculture are more food secure with a food security index (0.7564 and 7071) respectively, compared to their non-adopters' counterparts with an index of 0.4931 and 0.4602. The differences observed between the nutrition-sensitive agriculture adopters and non-adopters among the male and female farming households could be attributed to the nutrition-sensitive agriculture adoption and how well they were able to cope and adapted to the food insecurity crisis during the period of inadequate food supply. The implication is that the impact of nutrition-sensitive agriculture adoption on household food security is felt more among adopters in both category than the non-adopters.

Table 10: Composite food security index of farming households

Food Security Indicators	Male farming households		Female farming households	
	Adopters	Non-adopters	Adopters	Non-adopters
Food Consumption Score (FCS)	0.6842	0.4498	0.6221	0.3629
Household Dietary Diversity Score (HDDS)	0.4925	0.3541	0.3898	0.3114
Months of Adequate Household Food Provisioning (MAHFP)	0.3639	0.2868	0.2767	0.1863
Coping Strategy Index (CPI)	0.2894	0.2662	0.2461	0.1479
Household Food Insecurity Access Scale (HFIAS)	0.4948	0.3314	0.3354	0.2525
Food Security Index (FSI)	**0.7564**	**0.4931**	**0.7071**	**0.4602**

Source: Field survey (2022)

Level of food security among male and female farming households

The results in Table 11 present the distribution of farming households by their level of food security in the study area. The food security index so computed lies between 0 and 1, with 1 indicating maximum food security and 0 indicating no food or food insecurity at all. Based on results in Table 11 majority (72.0 and 69.6%) of the male and female farming households who had adopted nutrition-sensitive agriculture categorized as food secure while only 38.7% of male and 40.8% of female under non-adopter category are food secure by all the indicators investigated. Also, some 28.0% of male and 30.4% of female adopters and most (61.3% of male and 59.2% of female) among the non-adopters, respectively failed to meet all the required indices of food security and thus, categorized as food insecure. This finding suggests that majority of farming households (male and female) who have adopted nutrition-sensitive agriculture are more food secured compared to their non-adopters' counterparts. The implication is that, adoption of nutrition-sensitive agriculture has raised the household food security of adopters more than the non-adopters.

Table 11: Distribution of farming households by their level of food security

Categorization	Food Security Index (FSI)	Male farming households				Female farming households			
		Adopters		Non-adopters		Adopters		Non-adopters	
		(f)	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*	*(f)*	*(%)*
Food secure	0.50 - 1.00	67	72.0	58	38.7	48	69.6	49	40.8
Food insecure	0.0 - 0.49	26	28.0	92	61.3	21	30.4	71	59.2
Total		**93**	**100**	**150**	**100**	**69**	**100**	**120**	**100**

Source: Field survey (2022)

Relationship between male and female farming households in their level of food security

Table 12 reveals that there was no statistically significant difference in the level of food security between male and female farming households who had adopted nutrition-sensitive agriculture in the study area. The

t-test values were 2.6172 and 2.5086 for male and female farming households, respectively at a p-value of 0.329. Results show that male respondents had a mean value of 15.031 and a standard deviation of 0.4392 while female respondents had a mean value of 14.976 and a standard deviation of 0.4146. Our findings suggest that no significant difference existed in the level of food security among the male and female farming households who had adopted nutrition sensitive agriculture. More findings in Table 12 show that the mean difference for male and female farming households was 0.055 which could have accounted for the significant difference in their level of food security.

Table 12: Results of independent sample t-test showing differences in the level of food security among adopters (male and female) farming households

Variable	N	Mean	SD	SE	MD	t-test	*p-value*	Decision
Male	93	15.031	0.4392	0.0125	0.055	2.6172	0.329	No significant
Female	69	14.976	0.4146	0.0131		2.5086		

Source: Field survey (2022); $p \leq 0.05$

Conclusion

Using an indicator-based approach, this study empirically investigates if the adoption of nutrition-sensitive agriculture could help in narrowing the gender gap in household food security in southwest Nigeria. Through a cross-sectional survey of farming households, we first assessed the determinants of adopting nutrition-sensitive agriculture and found that household size, total farm size, membership of association, access to extension services, awareness of nutrition-sensitive agriculture, and access to training tend to positively and significantly influence nutrition-sensitive agriculture adoption among male and female farming households, while access to extension did not influence adoption among female farming households' decision to adopt nutrition-sensitive agriculture. Similarly, using a robust food security model we found that there was no statistically significant difference in the level of food security among male and female farming households who had adopted nutrition-sensitive agriculture in the study area. However, the level of food security was higher among the nutrition-sensitive agriculture adopters than that of non-adopters in both categories of farming households and this was evident across all indicators of food security investigated thus, indicating the influence of adoption of nutrition-sensitive agriculture on household food security. Therefore, interventions that are aimed at improving the awareness of nutrition-sensitive agriculture among rural farming households should be encouraged, narrow gender gap in households' food security and may actually have significant influence on the adoption. Since, adoption of nutrition-sensitive agriculture has improved household food security among the adopters compared to non-adopters. This indicates that, if all the male and female farming households in a given region adopted nutrition-sensitive agriculture, their household food security would improve significantly. This suggests the potential of nutrition-sensitive agriculture to achieve improved households' food security and help to narrow gender gap in food security, pointing to the need for reaching the current non-adopters. Finally, further studies should be conducted to test the composite Food Security Index (FSI) in other areas using similar indicators or together with other indicators not included in this study similar as Household Hunger Scale (HHS), Self-assessed food security (SAFS).

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An Assessment of the Effects of Covid 19 Lockdown on Crop Farmers' Anxiety Disorder in Nigeria

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Abstract

The study investigated on an assessment of the effects of covid 19 lockdown on crop farmers' anxiety disorder in Nigeria. Descriptive statistics, Double difference model, average treatment effect, generalized anxiety disorder scale and Oslo social support scales were the analytical tools used. The results confirmed that the covid-19 lockdown indeed affected the anxiety level of the farmers. The lockdown had a negative effect on their production and it is in turn reduced their income drastically. Age, income, farmer's experience, land size, access to credit, extension visitation, access to technical support from agricultural organizations, access to support from families and neighbours were found to have significant effect on anxiety level. It was also found that most of the farmers received support from their families and neighbours than they did from the government. The anxiety levels of farmers engaged in maize farming were found to be higher than those in cassava farmers due to the fact that the anxiety level has somewhat reduced before the harvest of cassava. The COVID-19 pandemic lockdown also created different anxiety levels in the farmers causing them to be unable to stop or control worrying. In conclusion, farmers were seriously affected by covid 19 lockdown. The study recommended that in order to reduce the anxiety levels of maize farmers, it is necessary to provide good storage facilities and government should provide agricultural support policies, including financial resources and technical assistance can be implemented for those farmers who are heavily affected by the COVID-19 outbreak. These farmers should be supported for early recovery and resilience to anxiety

Keywords: Covid-19, generalized anxiety disorder scale, Oslo social support scales

Introduction

In December 2019, the novel Coronavirus Disease (COVID-19) was first reported in Wuhan China. The disease spread quickly all through the world, causing the passing away of many individuals (Cortignani, Carulli & Dono 2020). The World Health Organisation (WHO) announced this plague to be a pandemic on 11th March 2020. The disease grew into a serious global health challenge killing over 100,000 people in 100 countries and was thereafter declared a pandemic by the World Health Organization on 11 March 2020. After spreading through East Asia, Europe, and North America in early 2020, the COVID-19 global pandemic started affecting countries in Africa and Latin America. Its rapid global spread has affected millions of people already vulnerable to food insecurity and malnutrition due to the effects of conflict and other disasters (Spitzer et al., 2006). With the largest population in Sub-Saharan Africa, and long-standing travel and trade links within Africa and to the rest of the world, it seemed inevitable that the pandemic would eventually reach Nigeria. At that time, various health and safety precautions have been taken in Nigeria in order to forestall the spread of the infection. The rapid spread of the coronavirus disease (COVID-19) in Nigeria has provoked the quick implementation of disease containment and other COVID-19 response measures.

This first case was announced in Lagos, which eventually became the virus epicentre in Nigeria, after which it began to spread throughout Lagos, Ogun State, and the Federal Capital Authority (FCT) area of Abuja. Since then, the coronavirus (COVID-19) pandemic has continued to disrupt life around the world, with a toll on human lives and economic activities (Patnaik *et al*., 2020). The arrival of the pandemic set off a chain of policy actions, including public health and education campaigns, fiscal and monetary measures, restrictions on large sections of the economy, and compensating measures in the form of social protection for poor and vulnerable people (Onyekwena and Amara Mma, 2020). With weak and fragile health infrastructure, Africa was considered a critical playground for the pandemic, and given Nigeria's current dwarfed health systems, response to the pandemic was a major source of concern with infected patients requiring admission into intensive care units (ICUs) for acute respiratory diseases and severe acute respiratory syndrome (SARS COV-2) pneumonia. Outside the health challenge, the pandemic has grim implications for Nigeria, especially as the lockdown affected the movement of goods, services, and humans (Ohia Bakarey & Ahmad 2020).

The outbreak of covid-19 pandemic has caused different economic problems and it has increased the anxiety of many farmers in the world especially in Nigeria. The federal government of Nigeria instituted a lockdown at the end of March. The lockdown came as a shock to crop farmers in Nigeria because of the fact that the restriction of movement was indiscriminately implemented thus making them unable to go to their farms especially in far distance locations. It has reduced the living standards and has greatly affected the mental health of many rural farmers. Although, these measures were taken to protect the public from the pandemic, the mental health of many people has been adversely affected as a result of both pandemic anxiety and restricted lifestyle imposed by measures to control the spread of the disease. Before the outbreak of the virus, rural farmers are already subjected to anxiety disorder because of different uncertainties like climate change, soil fertility, lack of inputs and lack of storage facilities. The covid-19 lockdown made rural farmers more anxious because of the different challenges they faced during the lockdown. Farmers also could not have access to transport facilities so they had to sell their produce cheaply to middlemen. They also faced the problem of increase in the price of agricultural inputs such as fertilizers, herbicides and seeds because the lockdown affected the import and transportation of such goods.

Considering all these challenges rural farmers encountered during the lockdown, the farmer's mental health was greatly affected. Most farmers became depressed because they could no longer meet up their standard of living. Studies that critically look into the effect of covid-19 lockdown on farmer's anxiety disorder especially in the study area are scanty. The study seeks to address the following objectives: to determine the socio-economic characteristics of respondents, to evaluate the impact of covid-19 lockdown on agricultural production of rural farmers, to examine the level of anxiety of rural farmers in the study areas during the covid-19 lockdown and to determine the effect of social support and social economic variables.

Methodology

This research was carried out in Kwara and Oyo states, Nigeria. The capital of Kwara state is Ilorin. Kwara is situated within the North central geopolitical zone of Nigeria. Kwara state shares boundary with the Republic of Benin and with five states in Nigeria. These states are Niger in the north, Oyo, Osun, Ekiti in the South, and Kogi in the East. The state lies approximately within latitude 8°.30'N, 5°00'E, with 16 Local Government Areas, a total land area of 36,825 km² and an estimated population of 3,192,900 (Ajadi *et al*, 2017). The rainy season begins towards the end of April and last till October. The dry seasons begin in November and end in April. The temperature of the state ranges from 33°C to 35°C from November to January and from 34°C to 37°C from February to April. The total annual rainfall ranges from 990.3mm to 1318mm. The rainfall exhibits double maximal pattern. Relative humidity ranges from 75% to 88% from May to October and 35% to 80% during the dry season (Ajadi *et al*, 2017).

Oyo state is located between latitude 6º55'– 8º45'N and longitude 2º50'–3º56'E respectively in south-western Nigeria, West Africa. Its shared border with the Republic of Benin, Kwara state, Osun state and Ogun state on the west, north, east and south respectively. With a projected population of 7,976,100 in 2023 Oyo state is the fifth most populous in Nigeria. The state has a tropical climate type with distinct periods of wet and dry season. The mean Annual temperature is 21ºC while the annual rainfall ranges from 1000 mm to 1500 mm. The Wet season is between 230 to 260 days/year. Most soil in Oyo state belongs to the savannah group of soil (Poudel & Subedi (2020). Even during the rainfall months, average temperatures are between 24°C and 25°C, while annual range of temperature is about 6°C. Rainfall figures over the state vary from an average of 1200mm at the onset of heavy rains to 1800mm at its peak in the southern part of the state to an average of between 800mm and 1500mm at the northern parts of the State.

A three-stage sampling technique was employed. The first stage involved the random selection of three Local Government Areas in each state because virtually all local government areas are known for maize and cassava production. The second stage involved the random selection of four villages each from the three Local Government Areas in the two states. The third stage involved random selection of ten (10) respondents from each selected village. Forty respondents were sampled from each local government.

This study employed descriptive and inferential statistics base on the objectives. The analytical tools include:

1. Descriptive statistics
2. Inferential statistics which include: Double Difference Model, Generalized anxiety disorder scale (GAD-7) and Oslo social support scale (OSSS-3)

Descriptive Statistics

Descriptive statistics such as frequency, percentage, averages and means were used to describe the socio-economic characteristics of the farmers. Descriptive statistics was used to determine the socio-economic characteristics of respondents

Double Difference Model

The double difference analytical tool was used to measure the short term or long-term program effects or impacts on respondents (Verner and Verner, 2005). It is a quantitative method often used to estimate and compare changes in outcome before and after program for respondents (Chen *et al*, 2006). In order to use the estimator in question, there must be information on the respondents, all the individuals must be observed both before and after the program (Verner and Verner, 2005).

The baseline data used to provide data for 'before' the COVID-19 lockdown were the farmers' maize and cassava output production in the year 2019 while farmers' maize and cassava output production in the year 2021 were used to provide data for 'after' the COVID-19 lockdown. The year 2020 was used to provide data of their maize and cassava output production during the lockdown. The differences between the average output production of these farmers were estimated using the simple form of the Double Difference.

The simple version of the Double Difference model is specified as follows:

$$LC = \frac{(Y2019 - Y2020) - (Y2021 - Y2020)}{Y2020} \quad \text{Equation 1}$$

Where:

LC = COVID-19 Lockdown

Y2019 = Output production of maize and cassava before the lockdown

Y2020 = Output production of maize and cassava during the lockdown

Y2021 = Output production of maize and cassava after the lockdown

The Double Difference has the advantage of netting out the effects of additive factors that have fixed (time-invariant) impacts on output indicator. If the mean Double Difference in the output is positive, it indicates that the COVID-19 lockdown has a positive effect (increase) on the output of the respondents while if it is negative, it means the COVID-19 lockdown has not increased the output of the respondents (Spitzer et al., 2006; Simonyan *et al*., 2012; Wakawa, 2014). It was expected *a priori* that the respondents will have lesser outputs after the lockdown.

Generalized Anxiety Disorder Scale (Gad-7)

Generalized anxiety disorder (GAD) is a chronic and highly prevalent disorder in the adult population. Though it occurs at a substantial frequency, the rate of GAD diagnosis is low (Konkan. *et al.*, 2013). The Generalized Anxiety Disorder (GAD-7) Scale is widely used in clinical practice and research due to its diagnostic reliability and efficiency. The GAD-7 is reported to be a valid, brief test in clinical, investigational, and general population samples (Konkan. *et al.*, 2013 Johnson. *et al*., 2019).

This scale was used examine the level of anxiety of the respondents during the covid-19 lockdown with a 4-point Likert scoring system (0: not at all, 1: a few days, 2: more than half of the days, 3: almost every day). A cut-off value of 10 for the GAD-7 total score was determined to be the threshold value for a diagnosis of GAD.

Oslo Social Support Scale (Osss-3)

OSSS-3 consists of three items that evaluate the level of social support. It is used for epidemiological and population-based research. Study participants was asked three questions on how many close friends they have, how involved other people are in their lives, and the availability of help from their neighbours. The response categories were assessed independently for each of the three questions, and a sum score was created by adding the three scores. The Oslo Social Support Scale has been used in several studies, thus supporting its feasibility and predictive validity with respect to psychological distress (Kocalevent.*et al*, 2012; Obayori, Nchom & Yusuf (2020)). This study was in accordance with the principles expressed in the Declaration of Helsinki. Gazi University Ethics Committee Number is 2021-398.

Results and Discussion

Socio-economic Characteristics of Respondents

The result of socio-economic characteristics of respondents is presented in Table 1. The result shows that 73.33% of the respondents were males, 26.67% were females in Oyo state while 76 67% were males and 23.33% were females in Kwara state. It shows that farming in the locality is predominantly done by males. This might have profound implications on production efficiency. It also shows that the average age of respondents is 52 years in Oyo state and 56 years in Kwara state. This implies that the active age participates predominantly in agricultural production in the study areas. This might have implication on productivity in farming since the respondents of this age categories are expected to be energetic and perhaps to have gained great farming experience that can enhance their productivity. A further analysis of the result shows that they were still in their productive age and could still actively engage in farming. Distribution of respondents according to their marital status shows that majority of respondents were married in both states, 63.33% in Oyo state while 90% in Kwara state. This implies that there is a possibility to have more people in the household of the respondents that are married. In an African society, the amount of family labour available to an individual is closely related to the marital status of the household head and household size (Muhammad -Lawal *et al*, 2009). Also, all things being equal an average farmer first exhaust all sources of labour in his family before hiring labour in order to reduce cost of production. A lot of respondents 59.17% have primary education in Oyo state while more farmers have secondary education in Kwara state 40.00%. This implies that most of the farmers can read and write while some farmers cannot and this shows that they can easily have access to new innovations and techniques. Education is assumed to have positive influence on the adoption of improved technologies because its ability to obtain process, understand and

interpret the agricultural information coming to farmers from any direction (Bamire *et al*, 2002). This is also in line with Eric et al (2014) which stated that education is vital for improving agricultural productivity such that it helps give farmers better farming methods and keep them abreast with changing innovation and ideas. This table also shows the majority of respondents have income more than ₦300000 in the both states. This shows that most of the respondents have average reasonable income and their agriculture activities are successive.

Also, the results reveal that greater percentage (55%) of the respondents had farming experience above 15 years in study areas. This indicates that farming is an age-long venture to the respondents in the study areas. Over 57% belong to cooperative society while more than 42.50% are non-member of a cooperative society in two states. This implies that most of the farmers have access to credit from cooperative societies. Membership to a cooperative society has been founded to influence the adoption of agricultural technologies. It allows cross fertilization and fast transmission of ideas among farmers as implied by Akinola *et al*, (2010). Thus, this agrees with the findings of Owombo *et al*, (2011) who stated that, if the farmers have free access to credit facilities, they may be more interested in investing on improved technologies; therefore, it is expected to influence adoption of new technologies positively. Also, the result revealed that majority of respondents (91.67%) in Oyo state and (70.83%) in Kwara state did not have contact with extension agents during covid-19 lockdown while (8.33%) in Oyo state and (29.17%) in Kwara state have contact with extension agents during covid-19 lockdown. This suggested that during COVID-19 lockout, rural farmers have limited access to extension agents.

Table 1: Socio-Economic Characteristics of the Respondents (n= 120)

	Oyo State		Kwara State	
Characteristics	**Freq**	**Percent (%)**	**Freq**	**Percent (%)**
Gender				
Female	32	26.67	28	23.33
Male	88	73.33	92	76 67
Total	120	100.00	120	100.00
Age				
≤30	20	16.67	15	12.5
31-40	26	21.67	7	5.83
41-50	34	28.33	28	23.33
51-60	34	28.33	44	36.67
Above 60	6	5.00	26	21.67
Total	120	100.00	120	100.00
Marital Status				
Single	15	12.50	1	0.83
Married	76	63.33	108	90.00
Divorced	8	6.67	0	0.00
Widowed	21	17.50	11	9.17
Total	120	100.00		
Educational Level				
No Formal Education	7	5.83	29	24.17
Primary	71	59.17	31	25.83
Secondary	25	20.83	48	40.00
Tertiary	17	14.17	12	10.00
Total	120	100.00	120	100.00
Farm Size				
≤5	91	75.53	103	85.83
6-10	21	17.50	16	13.33

Above 10	8	6.67	1	0.83
Total	120	100.00	120	100
Farm Experience(years)				
1-5	19	15.83	11	9.17
6-10	19	15.83	17	14.17
11-15	16	13.33	27	22.50
Above 15	66	55.00	65	54.17
Total	120	100.00	120	100
Income (#)				
≤50000	3	2.50	10	8.33
51000-100000	6	5.00	8	6.67
101000-200000	10	8.33	12	10.00
201000-300000	6	5.00	30	25.00
Above 300000	95	79.17	60	50.00
Total	120	100.00		
Cooperative Membership				
Yes	69	57.50	71	59.17
No	51	42.50	49	40.83
Total	120	100.00	120	100.00
Extension Contact				
No access to extension	110	91.67	85	70.83
Access to extension	10	8.33	35	29.17
Total	120	100.00	120	100

Source: Field Survey, 2022

Effect of Covid-19 Lockdown on Crop Production of Maize and Cassava Farmers in Oyo and Kwara States

Result on the impact of COVID 19 lockdown on crop production is presented in table 2a. The result revealed that the farmers' productivity before, during and after covis-19 lockdown and differences within and between the years. The average productivity of maize production of farmers prior to covid-19 lockdown was 1421.23kg, and decreased to 820kg during the lockdown. The average productivity of cassava was 32120kg before covid-19 and reduced to 12214.23kg during lockdown. Likewise, it is also clear that once the COVID-19 lockdown was removed, the average output of maize production climbed from 820kg to 1326kg. Similar to how the average output of cassava production went from 12214.23 kg during the lockout to 16015 kg following the lockdown, as seen in Table 2. This suggests that as a result of the population being subjected to the COVID-19 lockdown measure, agricultural productivity, such as maize, has decreased by around 38.16%, while cassava production has decreased by about 23.73%. This could translate to retrogression in quest to half food security in the country which in turn led to spike in household food prices. This is well explicated by Gakpo (2020) who stated that the announcement of the lockdown had prompted food insecurity fears in Africa, which has triggered rising prices, panic buying and import-export disruptions. Libby (2020) revealed that COVID-19 lockdown had severely triggered scarcity and high prices of the main staple food beyond the reach of some people. In the same vein, Ojo (2020) noted that food shortages and price shocks for the consumers are key components of the lockdown crisis in Nigeria. All things being equal, spike in food prices reduces the quantity of food at household level, and low quantity of food is a potential threat to households' food security. This is because it limits the availability of food, which is a key component of food security. The foregoing was well captured in WFP (2020) report, which shows that COVID-19 lockdown measure is exacerbating food shortages in Nigeria. In table 2b, is presented

the result of the Double Difference impact analysis in Kwara state. It shows the average output(kg) of the farmers' maize and cassava production before, during and after the effect of the COVID-19 lockdown and the differences within and between these years. The result shows that the average output(kg) of maize production of the farmer before the COVID-19 lockdown was 1515.66kg. This decreased to 960kg during the lockdown. For cassava production, also the average output(kg) was 20330kg before the COVID-19 lockdown and decreased to 10174.17kg during the lockdown. This shows that the COVID-19 lockdown has caused a decrease in the output of the farmer's maize and cassava production.

However, it can also be seen that the average output of maize production increased from 960kg during the lockdown to 1294kg after the COVID-19 lockdown was lifted. Same as for the average output of cassava production which increased from 10174.17kg during the lockdown to 13905kg after the lockdown as shown in Table 2b. The percentage difference in maize output indicates that the COVID-19 lockdown had a negative effect of 25.81% on maize production. For the percentage difference in cassava output, as shown in Table 2b, it indicates that COVID-19 lockdown had a negative effect of 26.83% on cassava production. These resulted in hike in prices of maize and cassava in the market and low income to the farmers.

Table 2a. Effect of Covid 19 lockdown on farmers' productivity in Oyo State

	Before lockdown (kg)	**During lockdown (kg)**	**After lockdown (kg)**	**Percentage difference (%)**
Maize	1421.23	820.00	1326.00	**38.16**
Cassava	32120.00	12214.23	16015.00	**23.73**

Source: Field Survey Data, 2022.

Table 2b: Effect of Covid 19 lockdown on farmers' productivity in Kwara state.

	Before lockdown (kg)	**During lockdown (kg)**	**After lockdown (kg)**	**Percentage difference (%)**
Maize	1515.66	960.00	1294.00	25.81
Cassava	20330.00	10174.17	13905.00	26.83

Source: Field Survey Data, 2022.

Anxiety Level of Crop Farmers in Oyo State

The result anxiety level of crop farmers in Oyo State presented on Table 3 indicated that the GAD7 anxiety score of 46.67% of the respondents in the study area fell within 10-14 which means they have moderate anxiety, about 34% have GAD7 score ranged between 5–9 (were found to have mild anxiety); meanwhile 10.83% have score not more than 4(were found to have minimal anxiety) and 8.33% had between 15 – 21(were found to have severe anxiety), the average anxiety score in the study area was estimated to be 9.4. The result indicated that most of the respondents had moderate anxiety The Result was support by Rudolphi *et al*. (2020).

Table 3: GAD7score

Variables	**Freq.**	**Percent**	**Remark**
≤4	13	10.83	Minimal anxiety
5 - 9	41	34.17	Mild anxiety
10 - 14	56	46.67	Moderate anxiety
15 - 21	10	8.33	Severe anxiety
Mean = 9.4			
Total	120	100.00	

Source: Field Survey, 2022

Effect of Social Support and Anxiety Determinants of the Farmers in Oyo State

The result in Table 4 shows the effect of social support and anxiety determinants of the farmers during the covid-19 lockdown. From the result, it was deduced that most of the respondents(X=3.42) got motivational support during the covid-19 lockdown and it ranked first among the supports they were given. This is followed by family and relative support which ranked second (X=2.89) was also received by most farmers during the lockdown. Government palliatives ranked third (X=2.57) and technical support from agricultural organizations ranked fourth (X=1.71) which implies that few of the respondents got technical support

from agricultural organizations. This result implies that since most of the farmers did not get support from agricultural organizations, the anxiety of those farmers might be greater than those of the farmers who got support from agricultural organization. According to this result, farmers with strong social support had lower anxiety than farmers with less social support. The specific conditions of agriculture in every country require that this sector be protected and supported directly by the state or through institutions authorized by the state (Abay *et al*, 2017).

Table 4: Effect of Social Support and Anxiety Determinants of The Farmers in Oyo State

Variables	S.D F (%)	Disagree F (%)	Neutral F (%)	Agree F (%)	S.A F (%)	Total	Mean score	Rank
Technical support from agricultural organization	72 (60.0)	27 (22.50)	10 (8.33)	6 (5.00)	5 (4.17)	120 (100.0)	1.71	4th
Family and relatives support	17 (14.1)	28 (23.33)	33 (27.50)	35 (29.1)	7 (5.83)	120 (100.0)	2.89	2nd
Government palliative	32 (26.6)	37 (30.83)	13 (10.83)	27 (22.5)	11 (9.17)	120 (100.0)	2.57	3rd
Motivational support	16 (13.3)	12 (10.00)	32 (26.67)	26 (21.6)	34 (28)	120 (100)	3.42	1st

Source: Field Survey, 2022 (S.D= Strongly Disagree, S.A= Strongly Agree, F = Frequency)

Effect of Social Support and Anxiety Determinants of the Farmers in Kwara state

The result in Table 5 shows that the effect of social support and anxiety determinants during the COVID-19 lockdown. From the result, it was discovered that most of the respondents (M.S = 3.44) got support from their families during the COVID-19 lockdown and it ranked first among the supports they were given. This is followed by practical help from friends which ranked second (M.S = 2.89) was also received by most of the farmers during the lockdown. Technical support from agricultural organizations ranked sixth (M.S = 1.98) and government palliative support ranked seventh (M.S = 1.71) which implies that few of the respondents of the farmers got palliative support from the government. This result implies that since most of the farmers did not get support from the government, the anxiety of those farmers might be greater than those of the farmers who got palliative support from the government.

Table 5: Effect of Social Support and Anxiety Determinants of The Farmers in Kwara State

Variables	None Freq (%)	Rarely Freq (%)	Sometimes Freq (%)	Often Freq (%)	Always Freq (%)	Total	Mean Score
Technical support from agricultural organizations	46 (38.33)	39 (32.50)	28 (23.33)	6 (5.00)	1 (0.83)	120 (100.00)	1.98
People to count on, if there are great personal problems	28 (23.33)	39 (32.50)	37 (30.83)	12 (10.00)	4 (3.33)	120 (100.00)	2.35
Interest and concern people show	25 (20.83	32 (26.67)	37 (30.83)	21 (17.50)	5 (4..17)	120 (100.00)	2.58
Practical help from friends	23 (19.17)	23 (19.17)	34 (28.33)	24 (20.00)	16 (13.33)	120 (100.00)	2.89
Support from neighbours	28 (23.33)	16 (13.33)	33 (27.50)	29 (24.17)	14 (11.67)	120 (100.00)	2.88
Government palliative support	60 (50.00)	41 (34.17)	13 (10.83)	6 (5.00)	0 (0.00)	120 (100.00)	1.71
Family and relatives support	12 (10.00)	25 (20.83)	21 (17.50)	22 (18.33)	40 (33.33)	120 (100.00)	3.44

Source: field survey, 2022

Conclusion

The results confirmed that the covid-19 lockdown indeed affected the anxiety level of the farmers. The lockdown had a negative effect on their production and it in turn reduced their income drastically. Age,

income, farmer's experience, land size, access to credit, extension visitation, access to technical support from agricultural organizations, access to support from families and neighbours were found to have significant effect on anxiety level. It was also founded that most of the farmers received more support from their families and neighbours than they did from the government. The anxiety levels of farmers engaged in maize farming and were found to be higher than those in cassava farmers due to the fact that the anxiety level has somewhat reduced before the harvest of cassava. The COVID-19 pandemic lockdown also created different anxiety levels in the farmers causing them to be unable to stop or control worrying. These farmers should be supported for early recovery and resilience to anxiety. The study recommended that in order to reduce the anxiety levels of maize farmers, it is necessary to provide good storage facilities and government should provide agricultural support policies, including financial resources and technical assistance can be implemented for those farmers who are heavily affected by the COVID-19 outbreak

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Determinants of Adoption of Contract Farming by Nikki Soybean Producers, Benin Republic

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Abstract
In Benin, the enthusiasm of farmers to subscribe to contract farming is becoming more and more growing and raises questions and the attention of several actors. This article examined the factors determining the adoption of contract farming by soybean producers. Data were collected from 120 soybean growers using a systematic sampling questionnaire semi-structured and addressed to the farm manager. The data were analysed using STATA 15, and a probit model of adoption was estimated. The results show that the number of years of education (5%), the use of improved varieties (1%), the area available for soybean production (1%), contact with research services (10%) and exchanges with peers on contract farming (5%) issues positively influence the adoption of contract farming. Conversely, the existence of an off-farm activity (10%), the perception of agricultural risks (1%), the share of agricultural income in total household income (10%) and the distance between the producer's village and the main town of the commune (5%) had a negative effect on the adoption of contract farming. Thus, raising awareness of the advantages of contract farming and focusing on the characteristics of producers are factors that should be encouraged to adopt contracts.
Keywords: Contract farming, determinants, soybean, Nikki

Introduction
In Benin, soybean is one of the most widely grown crops because of its importance in reducing malnutrition and for a variety of uses (Kpenavoun *et al.,* 2018). Between 2009 and 2021, soybean production more than quadrupled from 57,000 ton to 253,954 ton (DSA/MAEP, 2022) and by 2030 an annual production of around 341,000 ton of soybean is forecast (AIEA, 2020). Increased soybean production not only contributes to food security and balanced diets, but also to the growth of the Beninese market. However, it should be noted that current yields obtained by producers remain low compared with the potential yield of 3t/ha for various recommended soybean varieties (ABS, 2021). Added to this are difficulties in accessing land, the market, specific inputs, inadequate and inappropriate storage infrastructure and poor mechanization of farming systems (MAEP, 2017). Similarly, producers' access to advice and agricultural credit remains very poor, with uncertainty over the prices at which products are sold (Aplogan *et al.,* 2022). In response to these constraints, farmers are adopting appropriate and innovative measures. Among the innovative solutions used by soybean growers is contract farming (Todomé *et al.,* 2019).

Contract farming is at the center of current debates about agricultural development options and the potential for developing the small-scale family production sector (Roy and Roy, 2017). It is an important governance mechanism, likely to improve the efficiency of value chains, and enables farmers excluded from formal financial systems to benefit from agricultural credit (Soullier and Moustier, 2018). It helps connect smallholders with buyers, shifting risk and the need for initial capital from small farming households (FAO,

2018; Arouna *et al*., 2021). It is also a source of production security and improved soybean quality, and leads to an increase in acreage (Ahoudjo, 2019; Todomé *et al.*, 2019). This helps to minimize the unpredictability of farming and to better plan agricultural production. It also facilitates access to the inputs needed to improve agricultural productivity and the marketing rate of finished products (Suwadu and Ibrahima, 2020). In addition, increasing productivity is essential for improving the livelihoods of small-scale farmers, who make up the bulk of the rural population. However, the process of adopting agricultural innovations is still very slow, and several aspects of adoption are still poorly understood, despite the fact that they are an important way out of poverty (Simtowe, 2011). Indeed, while it is true that contract farming has many advantages, the rate of participation of producers in the contract farming is not the same in the municipalities of Benin. The contract enforcement is said to be particularly difficult in basic food chains (Swinnen *et al.*, 2010). The adoption of contracts is a preferred choice of large farms in order to save on supply transaction costs (Weatherspoon and Reardon, 2003). According to Loevinsohn *et al.* (2013), farmers' decisions to adopt an innovation (contracting) are conditioned by the dynamic interaction between the characteristics of the technology itself and the range of conditions and circumstances.

The Beninese context is characterized by small family farms, in addition to growing soybean production with contract farming practices. It is therefore important to understand the mechanism of adoption of contract farming in the Beninese context. The aim of this research is to analyze the factors determining the adoption of contract farming by soybean producers in the commune of Nikki. Several theories have been developed to understand the factors involved in the adoption of an innovation by producers. One of these theories is that of the diffusion of innovations, pioneered by Rogers. Rogers (2003) explains that the variation in the level of adoption of an innovation can be explained by the perception of five attributes of the innovation. These are relative advantage (perceived usefulness); the compatibility of the innovation with the values of potential users, their past experience and their needs; the possibility of testing it; and the visibility and complexity of the innovation. Thus, the adoption of an innovation would be facilitated by the characteristics of the innovation, the nature of the social system, the decision making unit, the communication channel and the skill of the extension worker. Van Den Ban (1994) analyses other factors and concludes that farmers attach great importance to their sources of information. The innovator's initial cost, risk factors and institutional factors should also be taken into account when analyzing the determinants of adoption (Feder and Umali, 1993; Lindner, 1987). Moore and Benbasat (1996) have also identified image (degree of social improvement), the voluntary nature of technology adoption and social norms and pressures as important factors in the decision whether or not to adopt an innovation, which seems to be absent from Rogers' theory. Thus, the adoption of contract farming would be influenced by: the need for specific investments; the need to improve the efficiency of the supply chain by reducing costs; the need to establish close coordination, the perceived usefulness of contracts, producers' past experiences and their sources of information. Other main reasons for using contracts are risk transfer (insurance), alignment of incentives (Dubois, 2001), market attributes and farm characteristics (Guo and Jiang, 2007).

Methodology

Study Area

The data used in the study were collected in the commune of Nikki. The commune of Nikki with a population of around 180000, covers an area of 3,171 km^2 and is located in the Borgou department. It is bordered to the North by the commune of Kalalé, to the South by that of Pèrèrè, to the East by the Federal Republic of Nigeria and to the West by the communes of Bembèrèkè and N'Dali (Fig 1). The commune's climate is Sudano-Guinean, with a rainy season and a dry season. The soils are derived from the evolution of recent sedimentary formations and are generally favorable to agriculture. This commune was selected because it is one of the top five soybean-producing communes in Benin (DSA/MAEP, 2021) and the leading soybean-producing commune in Borgou, with production estimated at 23686.87 ton for the 2021-2022 season. The choice of this commune is also justified by the high level of agricultural contracting by

producers for fast-growing crops such as soybean. At the level of each village, a count of producers (contracting or non-contracting) was carried out. Systematic random sampling was then applied to determine the producers surveyed.

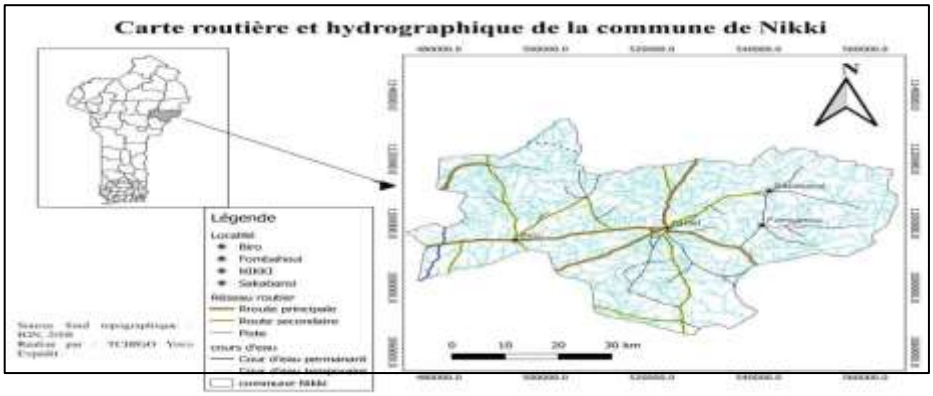

Figure: Map of the municipality of Nikki

Data Collected

Data were collected in three phases from 120 soybean growers in 4 villages. A documentation phase, during which theoretical explanations related to the topic were taken into account. An exploratory phase during which discussions were held with the various players involved in agricultural contracting and soybean production, where the questionnaire previously drawn up was tested in order to correct any errors. Finally, an in-depth phase during which the relevant data were actually collected.

Data Analysis

The data were analyzed using descriptive statistics (frequency, mean) and Inferential statistics such as logistic model for the adoption of contract farming. In the literature, two models are commonly used to determine the factors involved in adopting an innovation. These are the logit and probit models, which are similar and often lead to the same conclusion, and it is difficult to choose between the probit and logit models on theoretical grounds. However, probit is based on a standard normal distribution whereas logit is based on a standard logistic distribution (Sodjinou *et al.,* 2015). Both models are flexible and tend to perform well and provide parameter estimates that are asymptotically consistent.

In this study, the probit regression method was used given the qualitative and binomial nature of the dependent variable. This method has been proposed and used by several authors such as Belaidi (2013), Yabi *et al.* (2016) and Hountondji *et al.* (2018) in their study. The implicit model is expressed, thus:

$$Yi = f(X, e)$$

With, *Y*= dependent variable, the observed response of the i-th producer (i.e. the binary variable); Yi = 1 for adoption of the contract and Yi = 0 for non-adoption of the contract. *X*= matrix of variables likely to explain the variation in *Y* (table 1) *e*= distribution logistics error. The estimation of our probit model is based on the maximum likelihood method. The factors likely to influence the adoption of the contract by farms are presented in the table. Taking the variables into account, the empirical model can be written as follows:

Iij = δ0j + δ1jAGEij + δ2jGENij + δ3jDISTij + δ4jALPHij + δ5jNNIVij + δ6jCOOPij + δ7jPERij + δ8jSUPSij + δ9jEXTGij

+ δ10jSRESij + δ11jNEXPij+δ12jVULGij + δ13jUVAMij + δ14jEPAIij + δ15jPREVij + uij

Where ***j*** and ***i*** are the indices and u_{ij} the error term as defined in equation (1), the coefficients $\delta 0_j$ are the constant terms and δ_{ij} the parameters to be estimated which directly give the impact of the factor represented by the variable access to index-based agricultural insurance j.

The hypothesized effects of independent variable on the dependent variable is explained thus;

The age of the producer (AGE): this has a deterrent effect on the adoption of innovation.
When producers are older, they tend to be more wary of certain innovations, given their past experience. Moreover, flexibility and effort are associated with a preference for work (Kumar *et al.,* 2010). Also, as farmers advance in age, they become more skilled, thanks to learning by doing. Other authors like Maganga, (2021), believe that older producers may be more inclined to innovation than younger farmers. The expected sign is therefore negative.

Literacy (ALPH): Literacy sessions provide an opportunity for producers to share information and experiences. This enables producers to reduce uncertainties and gives them a more objective view of the innovation and the risks involved. A positive sign is therefore expected. ***Level of education (***NNIV): The level of education is recognized as favoring the adoption of innovation. Indeed, access to new information enables the most educated producers to better evaluate the innovation and thus limit their level of uncertainty (Roussy *et al.*, 2014). Moreover, producers' education plays an important role in understanding and implementing contract farming. It is therefore expected to be positive with the adoption of contract farming.

Share of farm income in household income (FREV): Off-farm diversification of household income also plays an ambiguous role on the adoption of new farming practices or technologies (Roussy *et al.,* 2014). Producers with additional off-farm income, reduce their need for financial support from traders. As a result, they adopt contract farming less. The plus minus sign is thus expected.

Contact with extension or research services (VULG/ SRES): the number of times the supervisor or extension agent visits the grower's field enables the grower to acquire knowledge in terms of know-how. This will enable growers to make the level of effort needed to produce a crop of the desired quality. A positive sign is therefore expected.

Distance between village and commune chief town (DIST): When the producers' village is very far from the commune chief town where most of the traders live, this will have a negative impact on the adoption of contracts. The further away the village is, the more the producer or trader spends on transport costs. The expected sign is therefore negative.

Perception of contract farming risk (PER): The perception of risk is one of the main factors in the rejection of innovation (Pannell *et al.,* 2005). In fact, some producers do not like taking certain risks, especially when they consider them to be too great. The expected sign is therefore negative.

Experience in soybean production (NEXP)*:* The role of experience in the adoption of an innovation is not clear. Indeed, experience facilitates the adoption of innovations that reduce perceived risk (Danquah *et al.,* 2013). The most experienced farmers are more likely to engage in contract farming (Maganga, 2021); in contrast, Tegene *et al.* (2000) found that the most experienced farmers have a shorter planning horizon, which does not encourage them to change their practices. The effect of experience on adoption is a function of perceived risk (Kebede *et al.,*1990). A plus-minus sign is therefore expected

Use of improved varieties (UVAM)*:* The use of improved seed at grower level has a positive effect on the adoption of contracts. Producers have access to improved soybean seed through production contracts. A positive sign is therefore expected.

Gender of producer (GEN)*:* Men have enough land and therefore sow a lot of area that requires the use of technology or innovation. In addition, women often have difficulties implementing innovations (Djinadou *et al.,* 2018). According to a study by Bellemare (2012), women may be less likely to adopt contract farming than men in Kenya, Madagascar and China respectively. So men are more likely to adopt innovations than women. A positive sign is therefore expected.

Exchange with other producers and membership of a cooperative (EPAI/COOP): Membership of a producer organization has a positive effect on the adoption of innovation. Belonging to a group facilitates access to technologies, especially recent ones (Kotchoni *et al.,* 2019). A positive sign is therefore expected.

Extra farm activities (EXTG)*:* Extra farm activities, through the financial resources they generate, facilitate the adoption of capital innovations (Gedikoglu *et al.,* 2011). But Entrena and Arriaza (2013), find that the plurality of activities is a brake on the adoption of innovations. The plus minus sign is therefore expected.

Area available for soybean production (SUPS): For farmers who own the land under cultivation, the decision to increase the area under maize only slightly increases the probability of adopting the technical package; whereas farmers who rent the land they cultivate are more likely to intensify the crops they grow on it and thus to adopt technologies that optimize yields (Mabah-Tene *et al.,* 2013). The plus minus sign is thus expected.

Results and Discussion

Socio-economic Characteristics of Respondents

The results on the characteristics of the adopters reveal that the majority of adopters are men (95%), with little education (33%), carrying out several agricultural activities (50%) and that the majority of these people (70%) have no schooling. Also, few of these producers have activities outside agriculture (50%). Most of these soybean producers belong to producer organizations. Over 65% of those surveyed belong to a soybean production cooperative. Access to research services was very low (21%) among all the producers. However, producers who signed storage contracts (42.86%) and marketing contracts (35%) had much greater access to research services than other producers.

Overall, the majority of contract growers take part in agricultural training (57%), on good soybean production practices (53%) and exchange knowledge with other growers (59%), unlike noncontract growers who receive very little agricultural training. Participation in meetings on agricultural contracting is low among contract growers as a whole, but high among adopters of storage contracts (35%), production contracts (36%) and marketing contracts (46%). In terms of the soybean seed used, contract growers make much greater use of improved soybean varieties than non-contract growers: 34% compared with 2% for all contract growers, around 59% compared with 2.5% for production contracts, almost 64% compared with 15% for storage contracts and 57% compared with 10% for marketing contracts. There is also a low level of use of improved seed among adopters of pre-financing contracts (33%).

Producers' perception of the risks associated with agricultural contracting varies from one form of contract to another. Overall, non-contracting producers perceive the risks of agricultural contracting to be much greater (56%) than contracting producers (30%). Producers' perception of the risks of contracting is much higher for production contracts (56%), storage contracts (74%) and pre-financing contracts (65%). As far as literacy is concerned, contracting producers have a higher level of literacy (43%) than non-contracting producers (32%). The distance between the village and the main town of the producers' commune also varies according to the status of the producer (contracting or non-contracting), but there is no difference in age between producers who adopt contract farming and those who do not. The share of agricultural income in household income was slightly higher (8.3/10) for contract farmers than for non-contract farmers (7.64/10). Finally, producers with the most arable land available for soybean production are the ones who adopt contract farming the most.

Table 3: **Socio-economic characteristics of respondents (n= 120)**

Contracts		Production	Storage	Marketing	Traditional financing	Package
Gender (%)	Female	5,13	0,00	2,17	3,70	4.29
	Male	94,87	100	92,86	96,30	95.71
Producer instructions (%)		30,77	35,71	32,14	40,74	32,86
Off-farm activities (%)		43,59	64,29	57,14	59,26	50,00
Membership of a cooperative (%)		97,44	85,71	100	59,26	65,71
Contact with research departments		25,64	42,86	35 .71	25,93	21,43
Soybean production training (%)		79,49	85,71	85,71	55,56	52,86
Exchange with peers (%)		76,92	85,71	75	64,81	58,57
Participation in meetings on soybean contract farming (%)		35 ,90	35,71	46 .43	25,93	21,43
Use of improved variety (%)		58,97	64 .29	57,14	33,33	34,29
Perception of Contract risk by producer		15,38	42,86	25	33,33	30,00
Agricultural training (%)		79,49	78,57	82,14	62,96	57,14
Literacy (%)		43,59	50,00	60,71	53,70	42,86
Distance between village and main town (km)		21,35	22,92	17,46	21,05	21,05
Age of producer (year)		39,4	42,9	42,6	40,8	40
Share of farm income in household income (x/10)		8,55	8,28	7,98	8,31	8,31
Area available (ha)		13,10	20,78	15,07	11,53	11,21

Source : Survey results, 2022

Factors determining the adoption of contract farming

The probit adoption models estimated for the different forms of contract farming are significant overall at the 1% level (P-value < 0.000). Marginal effects were also estimated. The results (Table 3) show that the factors determining the adoption of the different forms of contract farming are the distance between the producers' village and the main town, the existence of off-farm activities, the area available for soybean production, the number of years of education, the perception of the risk associated with contract farming, the share of farm income in total income, exchanges with other producers and the use of improved varieties.

Distance between the producers' village and the commune's main town

The distance between the producers' village and the commune chief town had a significant and negative effect at the 5% threshold on the adoption of marketing contracts. This implies that the further the producers' village was from the commune chief town, the more likely they were to adopt marketing contracts (0.8%). This result reflects the fact that most large traders are found in the towns (commune capitals), and with the poor state of access roads to villages, these traders find it difficult to travel to distant villages to sign marketing contracts. Also, the further away the village, the more expensive it is for traders to transport their goods, which reduces their profit margins. Traders wanted to maximize their profits, so they preferred to sign marketing contracts with producers in villages close to the main towns in order to make it easier to visit and inspect the products. According to Mounirou (2020), the cost of transport is proportional to the distance between the player's production site and the nearest market. The further away the village, the higher the transaction cost, which determines the level of rationality of the players (Arouna *et al.*, 2015).

Existence of off-farm activities

The existence of extra-agricultural activities is significant and negative with the adoption of production contracts at the 10% threshold. This implies that when the producer has several activities outside agriculture (trade, rearing, public service), the probability of adopting production contracts decreases by 33%. Thus, producers with no off-farm activities are much more likely to adopt production contracts. This can be explained by the fact that, for many producers, extra-agricultural activities constitute an important source of income, enabling them to draw on these extra-agricultural resources to support agricultural activities without having to resort to external financing in the process of producing and storing soybean. Also, when producers are able to buy inputs in cash, they find no reason to engage in contract farming (Maganga, 2021). Similarly, Entrena and Arriaza (2013) find that the plurality of activities is a brake on the adoption of innovations. But for Gedikoglu *et al.* (2011), off-farm activities, through the financial resources they generate, facilitate the adoption of capital innovations.

Area used for soybean production

The area available for soybean production has a significant and positive effect on the adoption of storage and marketing contracts at the 1% and 10% thresholds respectively. In fact, when the producer has a large area available for soybean production, the probability of adopting storage and marketing contracts is high. The influence of this factor on adoption can be explained by the fact that producers who have a large area for soybean production tend to plant a lot of land to soybean, which has a positive effect on their total production (*Ceteris parubus*). The adoption of storage and marketing contracts represents an opportunity for producers. In order to sell the quantities of soybean produced at a better price, most producers use storage/warranty and marketing contracts to secure their produce (against theft and dumping) while waiting for a better selling price, but also to benefit from financial support from traders while guaranteeing their sales market. Roussy *et al.* (2014) have also shown that farm size is an indicator of wealth for the farmer. However, it should be noted that increasing farm size requires additional labour resources. If the innovation is not easy to apply, the farmer may have difficulty implementing it on a large area (Anderson *et al.*, 2005).

Education level

The number of years of education of producers has a positive influence on the adoption of production contracts and traditional financing at the respective thresholds of 5% and 1%. Thus, the more educated the producer, the greater the probability of adopting production contracts and traditional financing. Indeed, with a higher level of education, producers are better informed about contract farming, which facilitates their adherence to contracts. According to Roussy *et al.* (2014), better-educated producers have more information at their disposal, enabling them to better evaluate the innovation and thus limit their level of uncertainty. According to Feder *et al.* (1995), the best-educated farmers are the first users of modern technologies and apply modern inputs more effectively throughout the adoption process. The level of education and literacy in local languages appear to be the only determining factors in demand (Mayoukou and Kertous, 2015).

Perception of contract farming risk

The perception of the risk associated with contracting has a negative influence on the adoption of production contracts at the statistical threshold of 1%. When producers perceive the disadvantages or measure the risks that may arise or that they incur by contracting for soybean production, they do not adopt contracts. In fact, some producers do not like taking risks, especially when they consider them to be too great. As the agricultural environment is characterized by a virtual absence of agricultural insurance, it would be difficult to adopt an organizational innovation where the certainty of profit is not perceived. Pannel *et al.* (2005) take a similar approach, explaining that risk is one of the main factors in the rejection of innovation.

Share of farm income in total income

The share of agricultural income in the total household income of producers has a significant and negative effect on the adoption of marketing contracts at the 10% threshold. This result implies that when the share of agricultural income in total household income is high, producers tend not to adopt marketing contracts. Thus, it is the producers who do not have enough agricultural income who tend to resort to marketing contracts in order to sell their soybean and meet family needs.

Contact with research departments

Producers' contact with research services has a positive and significant effect on the adoption of marketing contracts at the 10% threshold. This result can be explained by the fact that research service agents in the field make producers aware of the various opportunities (forms of contract) so that they are well equipped to make a success of their agricultural campaigns. The extension agents act as intermediaries between the innovators (researchers) and the users (producers) of the innovation. These results are in line with those of Mounirou (2020), who showed that the role of extension and research services in disseminating information is a key factor influencing the probability of adoption and the income of stakeholders. Extension agents are sources of information on agricultural innovations (Sodjinou *et al.,* 2015). This promotes the adoption of innovations by producers who have contact with them.

Exchange with their peers

Exchanges with other producers have a positive influence on the adoption of traditional financing contracts at the 5% threshold. The more a producer exchanges with other producers, the more likely he is to adopt traditional financing contracts. This can be explained by the fact that during exchanges between producers, information and knowledge about the players involved in financing, the conditions of access (contracts) and experiences of agricultural contract are shared. This sharing of information between producers facilitates the penetration and adoption of agricultural contracting. It is therefore a highly effective tool for sharing knowledge and adopting innovations. According to Roussy *et al.* (2014), the acquisition of information and experience within groups reduces uncertainty and enables farmers to perceive innovation and the risks involved more objectively.

Improved soybean varieties

The use of improved varieties by growers has a positive and significant effect on the adoption of production, storage and marketing contracts at the respective 1% thresholds. The need for growers to improve their yields through access to improved seeds and for reasons of inadequate financial resources lead some growers to resort to production contracts in order to gain access to production inputs. At the end of production, products are stored in warrantage warehouses under the storage contract to prevent them being sold off. It should also be noted that some producers have access to improved soybean seed through production contracts. The adoption of contract farming and improved soybean seed are complementary innovations in this context.

Overall, the adoption of production contracts is positively influenced by the number of years of education and the use of improved soybean varieties, on the one hand, and by the existence of off-farm activities (10% threshold) and the perception of risks associated with agricultural contracting, on the other. The adoption of storage contracts is determined by the area available for soybean production and the use of improved varieties. As for marketing contracts, their adoption is positively influenced by factors such as the area available for soybean production, contact with research services and the use of improved varieties. On the other hand, factors such as the distance between the village and the commune capital and the share of agricultural income in total household income have a negative effect on the adoption of this form of contract. Finally, the adoption of traditional financing contracts is influenced by the number of years of education and exchanges with other producers.

Table 2: Determinants of contract farming

Codes	Independent variables	Production		Storage		Marketing		Traditional financing	
		Coef (Std.Err)	dy/dx (Std.Err)	Coef (Std.Err.)	dy/dx (StdErr.)	Coef. (Std. Err.)	dy/dx (Std.Err.)	Coef.(Std.Err.)	dy/dx (Std.Err.)
DIST	Distance between village and main town	-0,005(0,03)	-0,001(0,01)	0,024(0,05)	0,001(0,01)	-0,081(0,03) **	-0,008(0,01) ***	0,026(0,02)	0,006(0,01)
EXTG	Off-farm activities	-1,742(0,94) *	-0,329(0,17) *	1,753(1,32)	0,052(0,04)	-1,580(0,99)	-0,181(0,12)	0,487(0,74)	0,119(0,18)
AGE	Age of producer	-0,534(0,33)	-0,097(0,06)	-0,090(0,36)	-0,002(0,01)	-0,061(0,29)	-0,006(0,03)	0,177(0,20)	0,043(0,05)
SUPS	Area available for soybean production	0,08(0,06)	0,015(0,01)	0,120(0,04) ***	0,003(0,002) *	0,069(0,04) *	0,007(0,01)	0,005(0,03)	0,001(0,01)
NNIV	Number of years of education	0,210(0,08)**	0,038(0,01) ***	0,017(0,09)	0,001(0,01)	0,039(0,07)	0,004(0,01)	0,15(0,06) ***	0,03(0,01)***
ALPH	Literacy	-0,437(1,01)	-0,077(0,17)	-1,978(1,09)	-0,054(0,14)	1,200(0,76)	0,139(0,09)	0,647(0,57)	0,159(0,14)
PER	Perception of contract farming risk	-3,275(1,19) ***	-0,444(0,13) ***	1,334(0,92)	0,052(0,05)	-0,953(0,79)	-0,087(0,06)	-0,337(0,54)	-0,083(0,13)
PREV	Share of farm income in total income	-0,214(0,21)	-,039(0,04)	0,213(0,38)	0,006(0,01)	-0,434(0,24) *	-0,044(0,02) *	0,165(0,19)	0,041(0,05)
SRES	Contact with research départements	1,689(1,18)	,374(0,27)	1,444(1,07)	0,073(0,09)	1,624(0,85) *	0,252(0,17)	0,748(0,83)	0,184(0,19)
EPAI	Exchanges with other producers	0,624(0,72)	,115(0,13)	1,323(1,18)	0,044(0,04)	0,440(0,80)	0,046(0,08)	1,289(0,56) **	0,310(0,13)**
UVAM	Use of improved varieties	4,673(1,16) ***	0,820(0,08)***	2,143(0,97)**	0,128(0,10)	2,848(0,87) ***	0,489(0,17) ***	0,611(0,66)	0,151(0,163)
Constant		2,108(2,29)		-8,426(4,40) *		2,242(2,507)		-4,488(2,12)**	
LR chi2 (16) =		81,28		37,92		53,26		38,29	
Prob > chi2=		0,0000		0,0001		0,0000		0,0001	
Pseudo R^2 =		0,7888		0,4386		0,4085		0,2318	
Log likelihood=		-36,231		-24,269		-38,560		-63,433	

Source : Survey results, 2022

Conclusion and Implications

Analysis of the determinants of contract farming adoption shows that the factors characteristics of the individual, the environment and the production context affect farmers' adoption behavior. On the one hand, the adoption of contract farming is positively influenced by the number of years of education, the use of improved varieties, the area available for soybean production, contact with research services and exchanges with peers on contract farming issues. On the other hand, the existence of off-farm activities, producers' perception of the risks associated with contract farming, the share of farm income in total household income, and the distance between the producer's village and the commune chief town all had a negative effect on the adoption of contract farming.

Based on the main results of this research, and in order to improve farmers' adoption of contract farming and access to agricultural services, we propose that the public authorities should raise farmers' awareness of the advantages of contract farming. The State (Ministry, public and private institutions) should also promote producer education and facilitate access to local information and improved seeds by setting up an appropriate agricultural advisory system (access to research and extension services). Steps should also be taken to facilitate access to arable land and create favorable conditions for exchanges between producers on contract farming issues.

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Appendix

Table 4: Variables introduced into the adoption model and expected signs

Variables	Descriptions	Terms and conditions	Expected signs
AGE	Age of operator (years)	Continuous variable	-
ALPH	Literacy	Qualitative variable (1=yes; 0=no)	+
DIST	Distance between village and main town	Continuous variable	-
PER	Perception of contractualisation risk	Qualitative variable (1=yes; 0=no)	-
NNIV	Number of years at school	Continuous variable	+
GEN	Type of producer	Qualitative variable (1=Male;0=Female)	+
NEXP	Experience in soybean production	Continuous variable	+/-
FREV	Share of farm income in household income	Qualitative variable (1=yes; 0=no)	+/-
SUPS	Cultivable area for soybean	Continuous variable	+/-
SRES	Contact with research structures	Qualitative variable (1=yes; 0=no)	+
VULG	Contact with extension services	Qualitative variable (1=yes; 0=no)	+
COOP	Membership of a cooperative	Qualitative variable (1=yes; 0=no)	+
EPAI	Exchanges with other producers	Qualitative variable (1=yes; 0=no)	+
EXTG	Extra agricultural activities	Qualitative variable	+/-
UVAM	Use of improved varieties	Qualitative variable (1=yes; 0=no)	+

Source : Field results, 2022

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Effect of Different Levels of *Moringa Oleifera* Oil on Milk Quality and Quantity of Goat

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Abstract
Moringa oleifera oil, obtained from the leaves of the *Moringa oleifera* tree, is extremely rich in phytonutrients and antioxidants. This study evaluated the quality and quantity of milk produced by West African dwarf Goats (n = 36) who were fed a *Moringa oleifera* oil (MOO)-based diet. The experiment was arranged as a completely randomized design, and it lasted for 42 days. Dietary treatments were: basal diet + 0.0 g kg-1 MOO (A, control), basal diet + 0.25 g kg-1 MOO (B), or basal diet + 0.50 g kg-1 MOO (C). Goats were fed the experimental diets and watering *ad libitum*. Diets were isonitrogenous, but the ether extract content varied from 0.88 (control) to 7.78% (MOO-based diets). The parameters evaluated are milk yield, milk quality (fat, crude protein, true protein, Non-Protein Nitrogen, Casein Nitrogen, Lactose, energy corrected milk, Lactic acid, Density, Solids not fat, freezing point, ash content, pH and Conductivity) There were significant effects of MOO on dry matter intake, milk yield, and energy corrected milk (ECM). Feeding MOO significantly increased milk fat from 5.21% in A to 9.42% in C. Similarly, the milk protein content increased from 3.55% to 5.35, and the milk yield increased from 78.20 to 101.41 kg. The total solids content and solids-not-fat content were significantly enhanced by the supplementation. Conclusively, supplementation of MOO in the diet of lactating goats enhanced milk fat and other nutrient contents of milk without adverse effects on consumption, milk quality and quantity of West Africa dwarf goats.
Keywords: *Moringa oleifera* oil, milk composition, milk yield, lactating goats, proximate composition of feed.

1,0 Introduction
Goat milk and milk products are vital for proper human nutrition, especially for people with lactose intolerance from cow milk. It is also important in developing countries where cow milk production is rather low. Milk could be obtained from sheep, cows, goats, buffalo, and deer. However, the management and production of milk from goats could be less cumbersome than those of cows and other large ruminant animals. Hence, developing dairy goat farming will enhance the economic and social value of the rural populace. The importance of goat milk is well elucidated in the literature (Belewu and Adewusi, 2001; Belewu and Ayeigbusi, 2002). The economic and nutritional value of goat milk is recognized in many countries of the world, hence, it is known as the "poor man's cow " in India (www.boergoatsindia.com/goat-farming) and "wet nurse of infant" in the United Kingdom (http://en.wikipedia.org/wiki/Human-animal_breastfeeding).

The current trend in the dairy industry is the production of value-added dairy products that are rich in polyunsaturated fatty acids (PUFA) as well as conjugated linoleic acid (CLA), which could offer potential

health benefits to humans. It has been reported that the Moringa tree, too, has potential health benefits. This makes it a good candidate for fortifying milk. Moringa is native to the Indian subcontinent but has become naturalized in tropical and subtropical areas of the world. Such regional names know the tree as Benzolive, Drumstick Tree, Horseradish, Saijihan, and Sajna (Fahey, 2005). *Moringa oleifera* is a multipurpose tree, as almost every part of the tree can be used for food, medication, and industrial purposes (Khalafalla *et al*., 2010). The leaves and fresh green pods are used as vegetables by humans and are rich in carotene and ascorbic acid, with a good profile of amino acids (Makkar and Becker, 1996). The tree has the potential to improve nutrition, food security and foster rural development (Hsu *et al.,* 2006). Studies from other countries indicated that the leaves have immense nutritional value, such as minerals, vitamins, and amino acids (Anwar *et al*., 2007).

In Africa and Indonesia, Moringa leaves are given to nursing mothers with the belief that it increase lactation (NRC, 2006, Astuti *et al.*, 2007). Additionally, the oil contains a high level of monounsaturated fatty acids, up to an average of 76.73%. Oleic acid is the predominant fatty acid and accounts for 73.57% of the total fatty acids. Therefore, the use of *Moringa oleifera* oil (MOO) in the diets of goats may contribute to the search for alternatives to the dairy goat diet, mostly during the dry season, and enhance the nutritional quality of dairy products. It is necessary to also underscore the fact that the medicinal value of MOO will benefit the animal and could be regarded as more environmentally and animal welfare friendly. Therefore, the thrust of this study was to evaluate the effects of supplementation of MOO at different levels (0.25 g kg-1 and 0.50 g kg-1) on milk quality and quantity in lactating West African Dwarf does.

2.0 Materials and Methods

2.1 Acquisition and Processing of *Moringa oleifera* Leaves

Moringa oleifera leaves were obtained from Ijapo estate, Akure, Ondo state, Nigeria. The leaves were removed from the stem and dried in the air. The air-dried leaves were ground after which they were soaked for 24 hours in a sealed flat bottom flask containing petroleum ether (Belewu *et al.*, 2012). Every 5-hour interval, the flask was shaken well to ensure uniformity. After 24 hours, the liquid was poured off and the oil was extracted using a Soxhlet apparatus.

2.2 Location of Experimental Site

The research was conducted at the Animal Production pavilion of the Department of Animal Production, University of Ilorin, Nigeria.

2.3 Experimental Animals and Management:

Pregnant West African Dwarf goats (n=36) purchased within Ilorin metropolis were utilized for this study. Upon arrival, the animals were treated for ecto and endo parasites using Ivomec. After giving birth, the kids were allowed to nurse the colostrum for three days and then separated from the does when it was observed that the colostrum had ceased. The does were randomly assigned to three experimental diets (Table 1) in a completely randomized design (CRD) model and kept separately in individual pens with experimental feed and fresh water available continuously throughout the experimental periods. Animals were milked twice daily (0800h and 1700h) and the total milk yield per day was recorded.

2.4 Experimental Diet

The experimental diets consisted of a basal diet without Moringa oil inclusion (A), basal diet with 0.25g/kg MOO inclusion (B), and basal diet with 0.50g/kg MOO inclusion (C). Other ingredients in the diets remained consistent across the treatments. With the MOO diets, the oil was added to the formulated diet every morning.

Table 1: Composition of the basal diet

Ingredients	%
Cassava waste	53.00
Groundnut cake	10.00
Rice husk	35.00
Vitamin-mineral premix	1.00
Common salt	1.00
Total	**100.00**

2.5 Sample Analysis

Milk quality evaluation (fat, crude protein, true protein, casein protein, density, conductivity, solids non fat, lactose, temperature, freezing point, ash content, pH and percentage lactic acid in the milk) was conducted utilizing the Lactoscan milk analyzer at the Shonga Dairy farms Ilorin, Nigeria. Samples of the diet were analyzed for proximate composition (AOAC, 1990).

2.6 Energy corrected milk (ECM) kg/d was calculated as: milk production (yield) x 0.327 + 12.95 x % fat + 7.2 x % protein (Shirley, 2006).

Feed efficiency = $\frac{\text{Energy- Corrected milk produced (ECM)}}{\text{Dry matter intake}}$

2.7 Data Analyses

All data collected were analyzed using a completely randomized design model (Steel and Torrie, 1980). The means were separated using Duncan multiple range test.

3.0 Results and Discussion

3.1 Proximate Composition of the Experimental Diets

Addition of MOO in the diets of lactating goats showed a numerical increase (87.60 to 91.49%) in the percentage of dry matter content (Table 2) across the diets, with diet C having the highest value (91.49%). The crude protein and ether extract contents also increased across the diets. The highest crude fiber was reported for diet B, followed by diet C and the lowest was diet A. Addition of MOO in the diets of the lactating goats reported herein influenced the ether extract content of the diets (2.66 -7.78%) which agreed with the reports of Bouattour *et al*. (2008); Fernandes *et al. (*2008); Bernard *et al*. (2009); Mdeiros *et al*. (2014). The increasing ether extract of MOO-based diet might be due to the presence of lipolytic compound in essential oil and this makes essential oil to be a concentrated hydrophobic liquid.

Table 2. Proximate Composition Nutrient Intake of the Experimental Diet*

Parameters	% DIET A	DIET B	DIET C	
Dry Matter	87.60	88.15	91.49	6.32
Crude Protein	7.47	7.77	8.17	1.45
Ether Extract	0.88^{a}	2.66^{b}	7.78^{c}	8.02*
Ash	12.99	11.86	11.57	1.79
Crude Fibre	3.42	41.59	37.44	1.26
Total fatty acid	2.48^{a}	4.69^{b}	6.30^{c}	0.95*

***Means of six determinations**

3.2 Goat Performance

Dry matter and Crude Protein intake (Table 3) were higher for the Moringa oil based diets (B and C) compared to the Control diet (A). The increased feed intake of Moringa oil based diet could be due to the improved palatability which stimulate appetite. This supported the report of Ornaghi *et al*. (2017) who noted higher dry matter and other nutrient intake in animals fed essential oil in their diet.

3.3 Milk Production

Table 3 displays the milk production and composition. The increasing milk production mentioned here aligned with the findings of Chilliard *et al.* (2001) who observed an increase in milk production when cows were given fat supplements during mid or late lactation. Adding MOO to the diet resulted in a significant increase ($P<0.05$) in milk production (g/d) from 78.20 in the Control group to an average of 98.64 (B) and 101.41 (C). This increase was consistent with the findings of Chion *et al*. (2010) who reported that the presence of essential oils or their derivatives could enhance specific sensory and nutritional properties of dairy products, adding value to the product. However, Bouattour *et al.* (2005) found no significant difference in milk production of goats fed lipid-supplemented diets.

3.4 Milk Fat

As a result of MOO supplementation, milk fat content (5.21(A) vs 8.88 (B) vs 9.42% C; $p<0.05$) and fat production (4.07 vs 7.96 vs 9.55kg) increased (Table 3). The increase in milk fat content was a response to MOO supplementation. It is important to note that essential oils contain bioactive compounds that can act as multifunctional feed supplements, promoting growth, digestion, and lipid oxidation. This finding is consistent with the studies of Morand Fehr *et al.* (1987) and Bouattour *et al.* (2008) who used prilled fat and soybean oil, respectively, in the diets of goats during early lactation. The response observed in this study was higher than that in dairy cattle (Chilliard *et al.*, 2001) and dairy goats (Bouattour *et al*., 2008). The positive effect of MOO on milk fat content reported in this study could be beneficial in addressing inversion syndrome in the dairy industry, especially in cheese production. This issue was not observed in this study, possibly due to the addition of MOO in the experimental animals' diet.

3.5 Milk Protein

Milk total protein content was 4.75%, 5.53%, and 3.55% in diets B, C, and A, respectively. Conversely, milk protein production was 2.78kg in diet A, 4.26kg in diet B, and 5.61kg in diet C. Additionally, true protein content was higher for MOO-based diets B and C. The utilization of true protein instead of crude protein demonstrated a better reflection of the economic value of milk protein as well as the nutritional value of milk. It also enhanced the accuracy of payment testing or protein by eliminating sources of random error. The protein fractions (true protein, casein protein, Non- Protein Nitrogen (NPN), and whey protein followed a similar trend as the crude protein content (Table 3). The result reported herein was not in line with that of De Peters *et al.* (1987), and Casals *et al*. (1999) who fed fat-supplemented diets to cows. The variations might be due to the types of fat supplemented in their diet. However, the results of this study agreed with the results of Bouattour *et al.* (2008). This also supported the assertion that the negative effects of dietary fat on milk protein content seems to be unusual in dairy goats (Baldi *et al.*, 1992; Chilliard *et al.*, 2006).There was a significant difference ($P<0.05$) in the percentage of the MOO based diets (B and C) revealing an upward trend in the percentage of the milk quantity and quality in relation to milk from the Control animals. However, animals on Diet B had intermediate values of all milk components compared to those of Diet C and the Control with no addition of MOO.

3.6 Conductivity:

It is the ability of substance to conduct electric current. Measurement of conductivity can be used to detect early mastitis. Currently, mastitis is the most expensive disease on dairy farm . The mean values of the electrical conductivity reported herein fell between 3 and 5Ms/cm (Germano *et al.*,1982) Changes in the milk conductivity are due to the concentration of ions in milk (addition of water, sugar, protein, insoluble solids, and salt). The value noted in this study is an indication that the animals were not having mastitis or damaged udder tissues.

3.7 Milk pH:

The milk pH noted in this study fell within the recommended value of between 6.4 and 6.7. Lower pH is an indication that the animal is having leukocyte infection such as Marmites. The pH of the milk to be used

for cheese production should be of excellent value since the pH value contributes to the softness or hardness of cheese . pH value is also vital in butter manufacturing process and it also enhances the shelf life of milk products (http://www.buzzle.com/article/ph-of-milk.html). The results of the study showed that the milk was not soured and without any microbial infection which could have enhanced the pH to be acidic hence, the milk was safe for consumption.

3.8 Acidity

The acidity of milk to be used for cheese production should be of excellent value since the acidity contributes to the softness or hardness of cheese. Acidity is also crucial in the butter manufacturing process and it also extends the shelf life of milk products. The findings of the study indicated that the milk was not spoiled and without any microbial infection which could have increased the acidity to be acidic hence, the milk was safe for consumption.

3.9 Lactic Acids:

It is crucial as a food preservative, curing, and flavoring agent. It is also used for decontamination during meat processing. The lactic acid observed in this study (0.18 to .20) is an added advantage to various pharmaceutical industries.

3.10 Solids-not-fat:

It contains lactose, casein, whey protein and mineral/ash. It is crucial for the improvement of the texture of ice cream, gives body and chew resistance to fermented products. The higher values (12.69 vs 14.77 vs 9.48 control) of the MOO based diets is a welcome development as it will enhance the production of milk products (sweetened condensed whole, skimmed milk, frozen condensed skimmed milk, buttermilk powder and many more).

3.11. Lactose:

It aids in the absorption of calcium and phosphorus as well as support good intestinal bacterial growth. The beneficial bacteria aid in the prevention of diseases and digestion of lactose. The higher lactose content of diets B and C will aid in the development of healthy brain and nerves. It also shows that the two diets will yield more energy, and aid in growth and cell development.

3.12. Density of milk

The density of milk and dairy products depends on the composition of milk (water, protein, fat, lactose salts). The density of milk decreases as the temperature increases due to the fact that the volume of fat changes much more with temperature than the volume of water. The assertion was confirmed in this study. The density value of milk is used to check the standard of milk (quality of milk). The increase in the density of milk (diets B and C) could probably be due to the higher milk yield and total solids contents of such milk.

3.13. Total Solids:

The total solids content of milk in this study became more for the MOO-supplemented diets as compared to the Control diet (A). The better total solids became determined to steer the increase and development of sensory acceptability and decrease appreciably the syneresis. It additionally has more advantageous on yoghurt production (Mahdian and Tehrani, 2007). The better total solids content of the MOO-supplemented diets is an introduced benefit to dairy farmers. The total solids content of Diets B and C increased appreciably (21.57 and 24.19 respectively). The increase can be due probably to the increasing fat, protein, and different elements of milk in this study.

3.14. Ash:

This represents the mineral content material. The result of this study confirmed that diet C had the better mineral content than B and the least was A. The better ash/mineral content of MOO-supplemented diets

gave credence to its dietary quality, physicochemical and microbiological values (miracletrees.org/moringa-medicinal-dietary-values-html)

3.15 Feed efficiency:

The feed efficiency (FE) reported herein was expressed as: Energy Corrected milk (ECM) divided by Dry matter intake (DMI). Energy corrected milk accounts for the true protein content of milk (Shirley,2006). Goats consuming diet C were more efficient followed by goats on diet B and the poorest was diet A (Control diet). This suggested that Diets B and C will improve net income and decrease ability of negative influences on the surroundings (Shirley, 2006). The effect of diets on FE can be evaluated in this study due to the fact that animals had been housed within identical surroundings and given comparable control routines. Variation in feed efficiency will be because of variations in the milk yield, protein, and fats and the dry matter intake (DMI) throughout the diets (Shirley, 2006). Other nutritional elements include diet formulation, digestibility, nutrient content material, and nutrient balance (Shirley, 2006).

The better overall performance of essential oil in animal diet become associated with stronger palatability of feed ensuing in better feed intake. The better feed performance mentioned in this observation agreed with the submission of Singh (2020) that important oil enhanced feed performance and boost health of livestock animals

Table 3: Nutrient Intake and Milk quality and quantity of the Experimental Animals

Parameers	**Diet A**	**Diet B**	**Diet C**	±SEM
Dry matter intake (g/d)	600.13	640.33	800.67	10.56
Crude Protein intake (g/d)	44.82	49.73	82.52	5.92
Butter fat(%)	5.21	8.88	9.42	2.12
Butter fat yield (kg)	4.07	7.96	9.55	2.98
Density (g/l)	32.15[a]	42.56[b]	49.74[b]	5.82
Electrolytic conductivity(Ms/cm	3.42	3.64	3.49	0.09
Solids not fat (%)	9.48[a]	12.69[b]	14.77[b]	1.82*
Protein (%)	3.5	4.75	5.53	0.68
Protein yield (kg)	2.78	4.26	5.61	0.33
True Protein (%)	3.42	4.58	5.33	0.18
Casein Protein	2.94	3.94	4.58	0.12
Non Protein nitrogen	0.13	0.17	0.20	0.02
Whey Protein	0.48	0.64	0.75	0.03
Lactose (%)	5.01	6.68	7.79	0.95
Lactose yield (kg)	3.92[a]	5.99[b]	7.90[b]	0.55*
Temperature (^{0}C)	27.10	23.80	21.43	6.50
Ash (%)	1.03[a]	1.22[b]	1.42[b]	0.19*
pH	6.57	6.58	6.56	0.04
Lactic acid (%)	0.20	0.18	0.19	0.02
Total solids (%)	1469[a]	21.57[b]	24.19[b]	2.12*
Milk quantity (g/day)	78.20[a]	89.64[b]	101.41[b]	15.37*
Energy Corrected milk (ECM)/d (Feed efficiency)	348.64	456.30	553.31	23.45
Energy Corrected Milk (ECM)/MI	1.74[a]	2.28[b]	2.34[b]	0.22*

Means having similar superscripts are not significantly differently from each other (P>0.05)

4.0 Conclusion and Recommendations

- The experiment strengthened a rational method to enhance milk yield and composition with the supplementation of MOO (an important oil). ·
- Feeding lactating West African dwarf goats with MOO-supplemented diets extended the fat, protein, milk yield, total solids, and solids not fat, lactose, and ash contents of milk. ·
- Supplementation of goat diet with MOO might be a possible and precious device for dairy farmers to provide milk and dairy merchandise wealthy in polyunsaturated fatty acids (PUFA) and conjugated linoleic acid (CLA) which can be useful to man. ·
- The consequences acquired from this study confirmed that the inclusion of MOO at 0.50g extended the milk amount and quality.
- However, in addition, studies are likewise encouraged to recognize the premiere degree of inclusion of MOO in the diets of goats.

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Effects of Postharvest Salicylic Acid Treatment on Some Quality Parameters of Sour Cherry (Prunus cerasus) Fruits during Cold Storage

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Abstract
The rising demand for sour cherry fruit is attributed to its nutritional benefits and economic significance. Given its perishable nature, preserving its quality and extending storage life are crucial for ensuring a sustainable supply of fresh fruits to consumers and processors. The current study examined effects of post-harvest salicylic acid (SA) treatments on quality parameters (respiration rate, soluble solid contents (SSC), titratable acidity (TA), total bioactive compounds such as phenolic contents, anthocyanin contents and antioxidant activity, and weight loss) of sour cherry 'Kütahya' variety during cold storage and shelf life period. In this research, three treatments (0, 1 & 2 mM salicylic acid concentrations) and three replications in completely randomized design were used. Mature harvested sour cherries were treated for 10 minutes at room temperature, thereafter stored at 0±1°C and 85-90% relative humidity for 42 days. Fruit quality parameters were assessed on weekly basis in the fruit taken at each analysis date and retained at 20 ± 1°C for 1 and 4 days. SA resulted in lower respiration rate (25.41 mL CO2 kg−1 h−1 for 1 mM SA), decreased TA (2.204±0.028 % malic acid mL for 2 mM SA). Both 1 and 2 mM SA significantly increased the fruit total phenolic contents (279.80±10.50 and 288.78±9.71 mg GAE/100 mL, 1 and 2 mM SA respectively) until 35th day, total anthocyanin contents (26.53±0.60 and 28.87±0.56 mg C-3-RE/ 100 mL, 1 and 2 mM SA respectively) and antioxidant activity (90.89±0.17 and 89.80±0.34 % inhibition, 1 and 2 mM SA respectively) until 28th day. Since SA applications, especially 1 mM SA, reduced respiration rate, decreased TA, and increased bioactive compounds, it could be considered as promising practice.
Keywords: Sour cherry, Quality, Storage, Shelf life, Salicylic acid, bioactive compound

Introduction
Sour cherry (*Prunus cerasus)* is one of important fruit with high commercial and human health potential benefits (Ferretti et al., 2010; Serradilla et al., 2017). It is well recognized for being good source of nutrients and bioactive compounds, including organic and inorganic substances, vitamins, dietary fibers, aromatic ad phenolic compounds substances particularly anthocyanins, quercetin, carotenes, and melatonin hormone (Pedisic et al., 2007; McCune et al., 2011).

These bioactive substances provide antioxidant capacities that aid in scavenging oxygen free radicals and lowering the risk of various chronic inflammatory illnesses, including Alzheimer's disease, cancer, cardiovascular disease and diabetes (Ferretti et al., 2010; McCune et al., 2011; Kelley et al., 2018; Blando and Oomah, 2019). The cherry consumption is also thought to enhance sleep, cognitive function, and pain relief following energetic activity (Kelley et al., 2018). The acidity and flavour of sour cherries make them highly appreciated and more appealing cherry products, and they are consumed dried, frozen, in liquor, wine, juice, and fresh in combination with other different fruit. (Aslantas et al., 2016). Thus, to ensure the

sustainable supply of fresh fruit, the producers need not only to increase the production but also to use appropriate postharvest technology to preserve the nutrient content of their product.

Nowadays, preharvest and postharvest application of the plant growth regulators are being used to facilitate in keeping postharvest quality of different fruits (Giménez et al., 2016). One of these growth regulators, salicylic acid, is a naturally occurring substance produced by plants that is safe for human health and plays a variety of crucial physiological roles, including controlling plant growth and development and boosting plant tolerance and resistance to different biotic and abiotic stress conditions (Hayat et al., 2010). Several studies showed that the pre and postharvest application of salicylic acid contribute to preserving quality parameters with prolonged storage and shelf life of different fruits (Valero et al., 2011; Cao et al., 2013; Giménez et al., 2015; 2016). However, there is no or limited information on effects of post-harvest salicylic acid treatment on extending post-harvest life and maintaining quality parameters of sour cherry fruit.

Hence, this study was designed to investigate the potency post-harvest application of various concentrations of salicylic acid on shelf life and some quality parameters (respiration rate, soluble solid contents, and titratable acidity, total phenolic content, total anthocyanin content and antioxidant capacity, and weight loss) of sour cherry 'Kütahya' variety under cold storage.

Material and Methods

Study Site

This study was conducted at Postharvest Physiology Laboratory of the Department of Horticulture, Faculty of Agriculture, Ankara University located in Ankara province, Türkiye at 39.950° E longitude, 32.860° N latitude, and an altitude of 857 meters with weather and climate characterized by hot, dry summers that may be fairly warm, with temperatures often exceeding 30°C and cold winters that can be chilly, with temperatures frequently falling below zero.

Fruit sampling, Treatments and Storage

In the study, "Kütahya" variety of sour cherry fruit (*Prunus cerasus*) harvested at the commercial harvesting time were used. The fruit were taken from the farmer's garden located at Çubuk district of Ankara province and brought right away to the Postharvest Physiology Laboratory. After selecting the fruit suitable for the experiment, salicylic acid (SA %0.01 Tween-20) was applied at different (0, 1 & 2 mM) concentrations with three replications by dipping method for 10 minutes at room temperature (20±1°C). The control groups were stored without any treatment. Thereafter, the fruit were preserved at 0±1°C temperature and 85-90% Relative Humidity (RH) for the period of 42 days.

Assessment of quality parameters

In this study, analysis was done on a weekly basis. On each analysis date, the quality parameters of the fruit taken from cold storage and retained at room temperature for the period of 1 and 4 days were assessed as follow:

Respiration rate

The respiration rate (mL CO_2 kg^{-1} h^{-1}) was calculated using 25 fruit samples kept at 20°C in a gas-tight jar for an hour and the amount of CO_2 in the jar was determined in percentage with a Servomex brand CO_2 analyzer.

Total soluble solid content (SSC) and titratable acidity (TA)

Total soluble solid content (SSC) was determined using a digital table refractometer and expressed as a percentage of Brix, whereas titratable acidity (TA) was determined by titrating a 50 mL mixture of distilled water and 1 mL of fruit juice with 0.1 N NaOH solution until pH equals 8.1 in an automatic titrator (Mettler Toledo DL 50 Graphix) and expressed as a percentage of malic acid. (AOAC 2012).

Bioactive compounds (phenolic contents, anthocyanin content and antioxidant)

The fruit total phenolic content was determined by spectrophotometer apparatus using the Folin-Ciocalteu reagent according to the method used by Rimpara et al. (2007) and Singleton and Rossi (1965) with minor modifications and the results were represented in mg gallic acid equivalent (GAE)/100 mL of fruit extract. The fruit extract was prepared by squeezing the cherries and thereafter 4 mL of distilled water was added to 1mL of obtained extract. The cherry extract was diluted again at 1/10 ratio and 1mL of this solution and 5mL of diluted Folin-Ciocalteu reagent at 1/10 ratio were mixed. This mixture was left for about 10 minutes and then 4 mL of Na2CO3 (75 g / L) was added and kept at room temperature. After an hour, the absorbance was determined at 765 nm wavelength. Total phenolic content was measured as gallic acid equivalent by comparing it with a standard curve created using 0, 25, 50, 100, 150, 200 and 250 ppm of gallic acid.

The total anthocyanin level was determined using the pH differential technique according to the methods used by Rimpara et al. (2007) and Proctor (1974) with minor modifications. The results were expressed as mg cyanidine-3-rutunozide (C-*3*-RE)/100 mL fruit extract. Briefly, the buffer solutions of 0.025 M potassium chloride (KCl) at pH 1 and 0.4 M sodium acetate ($CH_3CO_2Na.{}_3H_2O$) at pH 4.5 were used. The mixtures of 1mL diluted fruit extract and 4 mL of KCl or $CH_3CO_2Na.{}_3H_2O$ were kept at room temperature for approximately 30 minutes and the absorbance of the mixtures were recorded by reading the spectrophotometer at 520 and 700 nm wavelengths and Total anthocyanin was calculated as follows:

Total anthocyanin= (A x MW x Df x 1000)/ MA x C and A= (Amax-A700) at pH 1- (Amax-A700) at pH 4.5, where, Amax: Absorbance values at 520 nm as maximum wavelengths of cyanidin-3-rutinoside; A700: absorbance value at 700 nm is subtracted as there are other phenolic compounds present (Filimon et al., 2011); MW: Molecular weight of anthocyanin to be taken as a base (595.2 for cyanidin-3-rutinoside); Df: Dilution Factor; MA: Molar Absorption coefficient (28800 L/cm mg for cyanidin-3-rutinoside) and C: Thickness of the used cuvette (1 cm).

The antioxidant capacity was determined using the 2, 2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging activity according to Brand-Williams et al. (1995). The 60 µL of diluted fruit extract (1/5 ratio) and 1940 µL DPPH solution were mixed and absorbance of sample and control (methanol in place of fruit extract) at 517 nm wavelength were recorded and the antioxidant capacity was determined as follows:

Inhibition (%) = (Absorbance of control- absorbance of sample x 100)/Absorbance of control.

Fruit weight loss

The fruit weight loss was calculated using the method determined by Vasylyshyna (2018) whereby the percentage (%) of weight losses in fruit were calculated by porportioning the weight of the fruit at each analysis date to the initial fruit weight at the beginning of the storage.

Experimental design and statistical analysis

The experiment was performed in completely randomized design with three replications. Three way analysis of variance (ANOVA) of the obtained results at $P \leq 0.05$ was performed using MINITAB 17 package program. Significant differences were checked with Tukey test in MSTAT-C package program.

Results

Respiration rate

In this study, the storage period (SP), shelf life (SL), SA treatments (T), SP x SL and SP x T interactions were significantly affected in the sour cherry respiration rate (Table 1)

Table 1: Results of variance analyzes.

Assessments	Factors						
	SP	SL	T	SPxSL	SPxT	SLxT	SPxSLxT
Respiration rate (CO_2 kg^{-1} h^{-1})	0.000***	0.000***	0.000***	0.000***	0.046*	0.489 ns	0.643 ns
SSC (%)	0.000***	0.000***	0.000***	0.000***	0.000***	0.067 ns	0.000***
TA (malic acid %)	0.000***	0.000***	0.000***	0.000***	0.000***	0.038*	0.430 ns
Total phenolic content (mg GAE / 100 mL)	0.000***	0.000***	0.000***	0.000***	0.000***	0.007**	0.000***
Total anthocyanin content (mg C-3-RE/ 100 mL)	0.000***	0.968 ns	0.000***	0.038*	0.000***	0.130 ns	0.000***
Antioxidant capacity (% DPPH inhibition)	0.000***	0.028*	0.000***	0.351 ns	0.000***	0.006**	0.054 ns
Fruit weight loss (% and angle)	0.000***	-	0.580 ns	-	0.889 ns	-	-

[1]SP: storage period, SL: Shelf life, T: treatments (control, 0, 1, 2 mM SA), SP x SL: Storage period x shelf life interactions, SP x T: Storage period x treatments interactions, SL x T: Shelf life x treatments interactions, SP x SL x T: Storage period x shelf life x treatments interactions [2]*: P≤0.05; **: P≤0.01:***, P≤0.001; [3]ns, non-significant at P≤0.05.

The Respiration rate increased from the 7th to the 35th days of storage period and decreased in parallel with the prolongation of shelf life. The fruit treated with 1 mM SA had the lowest average respiration rate (25.41 mL CO2 kg^{-1} h^{-1}), whereas the highest average respiration rate (30.43 mL CO2 kg^{-1} h^{-1}) was recorded in the fruit treated with 2 mM SA (Table 2).

Total soluble solid content (SSC) and titratable acidity (TA)

In storage and shelf life processes, the SSC values changed depending on the effects of all factors (storage period, shelf life period and SA applications), except for the SL x T interactions (P=0.000). The TA values varied depending on the individual and interactive effects of all factors (P=0.000) (Table 1). While the SSC values increased in the first 7 days, it decreased as the storage period progressed after the 7th day of storage. The lowest SSC value was recorded in the controls and the highest SSC values were measured in other treatments. However, with the prolongation of storage and shelf life period, a decline in the TA values was recorded. Among the treatments, the highest mean TA value was measured in controls (2.408% malic acid) and the lowest value (2.204% malic acid) was measured in 2 mM SA application (Table 2).

Table 2: The changes in fruit respiration rate SSC, TA and bioactive compounds.

Factors	Respiration rate (mL CO2 kg^{-1} h^{-1})	SSC (%)	TA (%malic acid)	Total phenolic content (mg GAE/100 mL)	Total anthocyanin content (mg C-3-RE/ 100 mL)	Antioxidant capacity (% inhibition)
SP (day)[1]	P=0.000	P=0.000	P=0.000	P=0.000	P=0.000	P=0.000
0	26.06 ± 1.11 bc[2]	20.18 ± 0.18 c[2]	2.652 ± 0.037 a[2]	169.58 ± 3.15 d[2]	21.10 ± 0.32 d[2]	87.80 ± 0.52 c[2]
7	22.73 ± 1.04 e	23.06 ± 0.26 a	2.509 ± 0.019 b	220.95 ± 6.46 c	23.46 ± 0.54 c	88.55 ± 0.40 bc
14	25.02 ± 0.86 cd	22.90 ± 0.13 a	2.433 ± 0.022 c	227.41 ± 7.46 c	24.50 ± 0.79 c	89.51 ± 0.42 ab
21	22.97 ± 0.73 de	22.24 ± 0.09 b	2.439 ± 0.021 bc	231.20 ± 6.74 c	24.54 ± 0.66 c	89.82 ± 0.54 ab
28	27.94 ± 0.74 b	21.98 ± 0.12 b	2.265 ± 0.021 d	311.30 ± 13.00 b	31.18 ± 0.64 a	90.20 ± 0.38 a
35	35.45 ± 1.59 a	22.11 ± 0.08 b	2.102 ± 0.015 e	347.30 ± 12.50 a	28.01 ± 0.76 b	89.87 ± 0.30 ab
42	35.36 ± 0.82 a	22.22 ± 0.09 b	1.882 ± 0.014 f	316.01 ± 8.16 b	27.70 ± 0.59 b	90.30 ± 0.15 a
SL (day)	P=0.000	P=0.000	P=0.000	P=0.000	P=0.968	P=0.028
1	30.24 ± 0.88 a[3]	21.73 ± 0.09 b[3]	2.293 ± 0.018 b[3]	223.88 ± 4.18 b[3]	25.79 ± 0.44 ns	89.14 ± 0.22 b[3]
4	25.63 ± 0.53 b	22.49 ± 0.11 a	2.356 ± 0.020 a	297.18 ± 7.27 a	25.74 ± 0.39 ns	89.73 ± 0.23 a
T	P=0.000	P=0.000	P=0.000	P=0.000	P=0.000	P=0.000
Control	28.27 ± 1.10 b[4]	21.62 ± 0.17 b[4]	2.408 ± 0.028 a[4]	229.05 ± 7.53 c[4]	23.60 ± 0.51 c[4]	88.50 ± 0.38 c[4]
0 mM SA	27.62 ± 1.00 b	22.24 ± 0.15 a	2.335 ± 0.027 b	244.50 ± 7.55 b	24.00 ± 0.48 c	88.54 ± 0.27 c
1 mM SA	25.41 ± 1.02 c	22.20 ± 0.13 a	2.353 ± 0.023 b	279.80 ± 10.50 a	26.53 ± 0.60 b	90.89 ± 0.17 a
2 mM SA	30.43 ± 1.11 a	22.38 ± 0.13 a	2.204 ± 0.028 c	288.78 ± 9.71 a	28.87 ± 0.56 a	89.80 ± 0.34 b

[1]SP (day): Storage period, SL (day): Shelf life, T: Salicylic acid (SA) Treatments, [2] Range differences among storage periods,[3]Range differences among shelf life periods, [4]Range differences among treatments at P≤0.05.

Bioactive compounds (phenolic contents, anthocyanin content and antioxidant)

The total phenolic contents changed depending on individual and interactive effects of all factors (P≤ 0.05), whereas total anthocyanin contents changed depending on individual and interactive effects of other factors except SL and SL x T interaction (P≤ 0.05) and antioxidant capacity were significantly affected by individual and interactive effects except SP x SL and SP x SL x T interactions (Table 1). There was an increase in total phenolic compound contents from the beginning of storage to 35th day and on the 4th day of shelf life. The highest total phenolic contents were measured in the fruit treated with 2 mM SA (288.78 mg GAE/100 mL) and 1 mMSA (279.80 mg GAE/100 mL), and the lowest values were determined in controls (229.05 mg GAE/100 mL) (Table 2).

Total anthocyanin content increased from the beginning of storage until the 28th day of storage followed by a decreasing trend in the further storage period. Among the treatments, the highest fruit total anthocyanin content (28.87 mg C-*3*-RE/ 100 mL) was recorded in the fruit treated with 2 mM SA while the lowest values were measured in the control (23.60 mg C-*3*-RE/ 100 mL) and 0 mM SA (24.00 mL mg C-*3*-RE/ 100 mL) groups (Table 2).

The fruit antioxidant capacity showed an increasing trend during the storage period starting from the 7th day of storage and the highest values were found in the samples from the 28th (90.20%) and 42nd (90.30%) days. There was also an increase in the average antioxidant activity values depending on the prolongation of the shelf life period. Among the treatments, the highest average antioxidant value was found in fruit treated with 1 mM SA (90.89%), while the lowest values were recorded in the control (88.50%) and 0 mM SA (88.54%) groups (Table 2).

Fruit weight loss

The fruit weight loss had significantly affected by only storage period (P=0.000) (Table 1). It was increased with prolongation of storage period. Weight loss values were determined as 2.99% on the 7th day and 13.07% on the 42nd day of storage. None of the SA applications had a positive or negative effect on fruit weight loss (Table 3).

Table 3: The changes in fruit weight loss

Factors	Fruit weight loss (%)
SP (day)[1]	P= 0.000
0	0.00 ± 0.00 (0.00 ± 0.00) [2] g
7	2.99 ± 0.12 (9.93 ± 0.21) f
14	5.37 ± 0.11 (13.39 ± 0.15) e
21	7.53 ± 0.42 (15.87 ± 0.43) d
28	9.28 ± 0.54 (17.67 ± 0.49) c
35	11.07 ± 0.55 (19.38 ± 0.47) b
42	13.07 ± 0.57 (21.15 ± 0.46) a
T	P= 0.580
Control	7.51 ± 1.21 (14.19 ± 1.66) ns
0 mM SA	6.93 ± 0.95 (13.81 ± 1.49) ns
1 mM SA	6.69 ± 0.86 (13.65 ± 1.42) ns
2 mM SA	7.05 ± 0.92 (14.00 ± 1.47) ns

[1]SP (day): Storage period, T: Salicylic acid (SA) Treatments

[2] Range differences among storage periods, ns: non-significant at P≤0.05.

Discussion

Respiration Rate

Our results presented that the use of 1 mM SA delays the increase in sour cherry fruit respiration rate during cold storage and shelf life period. However, the high SA dose is not effective in reducing sour cherry

respiration rate. Different other studies in sweet cherries (Giménez et al., 2016); plums (Luo et al., 2011) and pomegranates (Sayyari et al., 2011) showed the similar results whereby SA application significantly reduced fruit respiration rate during cold storage. The low respiration rate in fruit means slow physiological mechanism and delayed fruit aging and senescence which leads to the preservation of organic compounds (Giménez et al., 2016). During the storage period salicylic acid, as a potential inhibitor of ethylene production and activity, can prevent the dramatic increase in fruit respiration rate by delaying the breakdown of starch into glucose (Asghari and Aghdam, 2010).

Total soluble solid content (SSC) and titratable acidity (TA)

The SSC/TA ratio determines the flavour and taste of fruits (Blando and Oomah, 2019). In the postharvest period, the fruit total soluble solid increases and titratable acidity decreases leading to an increase in fruit sweetness (Ferretti et al., 2010; Giménez et al., 2016). SA applications increase the levels of sugar, organic acid and total soluble solid content in various fruit (Sayyari et al., 2011; Razavi et al., 2014; Giménez et al., 2017; Sabır et al., 2019). Valero et al. (2011) also stated that sweet cherry fruit treated with 1 mM SA, ASA or oxalic acid (OA) had the lower titratable acidity than controls during cold storage. Our study showed the similar results whereby the fruit treated with 2 mM SA and 1 mM SA recorded higher SSC and lower TA values than the fruit in other treatments.

Bioactive compounds (phenolic contents, anthocyanin content and antioxidant)

The increase in the phenolic compounds of the fruit during cold storage is linked to the stress conditions from low temperature. Shah et al. (2018) stated that the polyphenol compounds, chlorophyll breakdown and color change of sour cherry fruit are likely to increase during postharvest period. The phenolic and anthocyanin compounds are recognised as beneficial antioxidants and have a scavenging effect on reactive oxygen species (ROS) (Asghari and Aghdam, 2010). According to different studies (Yao and Tian, 2005; Luo et al., 2011), salicylic acid boosts the fruit's ability to fight off reactive oxygen species (ROS) during storage by promoting the activity of antioxidant enzymes like phenylalanine ammonia-lyase (PAL), glutathione reductase (GR), peroxidase (POD) and polyphenol oxidase (PPO). The increased activity of PAL enzyme enhances the production of phenolic compounds (Guardo et al., 2013).

The high antioxidant capacity and high total phenolic and anthocyanin level in the fruit treated with salicylic acid (SA) are thought to be linked to the enhanced activities of these enzymes by the applied salicylic acid. Our findings agreed with the findings of Giménez et al. (2017) on ‘Sweet Late’ ‘Sweet Heart’, and ‘Lapins’ cherries and Giménez et al. (2014) on ‘Sweet Heart’ and ’Sweet Late’ cherry varieties stating that Acetyl salicylic acid (ASA) and salicylic acid (SA) applications increase antioxidant activity, total phenolics and anthocyanin contents. Valero et al. (2011) also found the increase in total phenolic and anthocyanin contents in the first 10 days of storage in the controls and the decrease in the first 10 days followed by gradually increase in sweet cherry fruit treated with 1 mM SA, ASA or or oxalic acid (OA) throughout the storage period.

Different other postharvest studies showed that SA applications increased antioxidant activity, total phenolic content and total anthocyanin levels in apricots (Ezzat et al., 2017), nectarines (Bal, 2016); plums (Davarynejad et al., 2015); peaches (Awad, 2013; Razavi et al., 2014) and kiwi (Fatemi et al., 2013). The same result was reported by Giménez et al. (2016) who reported that MeSA applications in 'Early Lory' cherry variety fruit increased the antioxidant capacity of the fruit while preserving the concentrations of total phenolic compounds and anthocyanins during storage period. García-Pastor et al. (2020) and Sayyari et al. (2011) also stated that postharvest ASA applications increase the total anthocyanin levels in pomegranates. Similarly, Koyuncu et al. (2019) also stated that pomegranate fruit treated with SA and putrescine had higher antioxidant activity and total phenolic compound content compared to controls.

Fruit weight loss

The increase in fruit weight loss in cherries during storage brings along with the physiological, biochemical and sensory changes (Shah et al., 2018). Our findings on weight loss are parallel to the findings of Baninaiem et al. (2016), who argued that SA applications do not have a substantial influence on tomato weight loss during storage. However, they are contrasted with the findings of the studies carried out on the nectarines (Bal, 2016); plums (Davarynejad et al., 2015); peaches (Razavi et al., 2014) and apples (Kazemi et al., 2011), The authors reported that SA applications during storage reduced fruit weight loss. This shows that the effect of SA applications on fruit weight loss in postharvest period varies by species. In addition, sour cherry fruit are very sensitive to water loss even under high relative humidity conditions and the use of packaging materials during storage should be taken into account while dealing with sour cherry fruit water loss.

Conclusion

The postharvest application of 1 mM SA delayed the increase in respiration rate and decreased TA levels. While the SSC increased in all applications during the cold storage, SA application significantly delayed this increase till 28 days of storage. Both SA concentrations (1 and 2 mM) increased total phenolic content in the first 35 days and total anthocyanin content and antioxidant capacity in the first 28 days of storage. Therefore, the postharvest SA applications, especially 1 mM SA application, can be used to preserve the quality of the sour cherry fruit 'Kütahya' variety for 28 days in the cold storage with 4 days of shelf life after the fruit are removed from the cold storage.

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Estrus Characteristics and Reproductive Hormone Production in Boran and Boran×Friesian Dairy Cows at Bishoftu Agricultural Research Center, Ethiopia

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Abstract
A study was carried out at the Bishoftu Agricultural Research Center to determine the blood hormone level and estrus behavior exhibited by purebred Boran and crossbred dairy cows from October 2019 to June 2020. Twenty multiparous dairy cows were purposively selected after gynecological and obstetrical examination, and blood samples were collected on days zero, 7, and 11 to measure serum concentrations of *estradiol* and P4. In addition, visual observation was carried out three times per day for the detection of estrous behavior. The result revealed that 35% and 30%, 10% and 25%, and 15% and 30% of the crossbred and Boran showed quiet acceptance to be mounted, disoriented mounts, and attempts or rejection to mount, respectively, during the study period. The mean estradiol level was 19.38 2.46 pg/ml on day zero, 16.08 2.16 pg/ml on day 7, and 15.46 2.09 pg/ml on day 11 (P<0.0001).The mean progesterone level was 0.310.08ng/ml on day zero, 1.120.32ng/ml on day 7, and 1.190.19ng/ml on day 7 (p 1.19±0.19ng/ml on day (p<0.05).The mean estradiol levels were 19.153.19 pg/ml, 16.211.49 pg/ml, and 15.42.14 pg/ml for good, medium, and poor BCS animals, respectively. where the progesterone levels were 1.1200.31ng/ml, 1.070.26ng/ml, and 0.6660.21ng/ml, respectively. Boranhad had significantly (P< 0.0001) higher (13.051.63 pg/ml) circulating estradiol concentrations than crossbred cows (20.891.75 pg/ml). The progesterone profile of Boran was significantly (p 0.0001) higher (1.120.23 ng/ml) than that of crossbred crossbred cows (0.99±0.26ng/ml).This difference is most likely due to the differences in metabolism and metabolic hormone concentration between Boran and crossbred cows. It is recommended that detailed research be done on the all-inclusive hormonal profile and the effect of the season and other management systems of the farm on the efficiency of reproduction.
Keywords: Boran, Boran × Holstein, Estradiol, Progesterone, Reproductive physiology

Introduction
Ethiopia has the largest livestock population in Africa [1]. Livestock production is an integral part of agriculture in the country and contributes about 16% of the GDP, over 40% to the agricultural GDP, 12–15% of the export earnings, 60–70% of employment opportunity [2, 3], and 37–87% of household incomes [4]. Ethiopia possesses the largest cattle population in Africa, estimated at about 65.35 million, of which 97.76%, 1.91%, and 0.32% are local, hybrid, and exotic breeds, respectively. Dairy cows are estimated to be around 7.15 million, and milking cows are about 12.57 million [5].

Despite the huge number of cattle and their economic importance, productivity remained very low due to many limitations. Among these are high disease and parasite prevalence, low quality and quantity nutrition,

low genetic potential of indigenous breeds, poor reproductive performance, limited access to veterinary and extension services, and poor husbandry and management practices, resulting in the poor reproductive performance of dairy cattle [6, 7]. Reproductive disorders have been found to be a major reason for decreased reproductive efficiency in cattle, and consequently, reproductive efficiency is the major determinant of the life-time productivity of cows [8]. In the last few decades, as the major epidemic diseases were brought under control, emphasis increasingly shifted to economically important diseases for dairy producers, and animal health problems stood out as the most prominent [7]. Regular breeding depends on the normal function of the reproductive system. In order to breed regularly, the cow has to have functional ovaries, display estrous behavior, mate, conceive, sustain the embryo through gestation, calve, resume estrous cyclicity, and restore uterine function after calving. Each of these aspects of reproductive function can be affected by management, disease, and the genetic make-up of the animal. When the function of the reproductive system is impaired, cows fail to produce a calf regularly [9].

Reproduction is a complex process whereby individuals generate their offspring to continue life [9]. The sexual activity of the adult female bovine, non-pregnant and in good health condition, is cyclic, and the cycle lasts for 20 to 21 days with extremes varying from 17 to 24 days; this is true for heifers and for cows [10]. The root cause of the declining health of dairy is a combination of a variety of physiological and management factors that have an additive effect on reproductive efficiency. The dairy industry depends on the reproductive performance of their dairy cows in order to meet the growing demand for products, and the lactation cycles are dependent on the cows ability to become pregnant since the hormones released during and after pregnancy are essential for the development of the mammary gland and increase milk production [11].

Reproductive efficiency is dependent upon optimization of management, wellbeing, and the physiology of cows. Although many factors are interrelated with the reproductive efficiency of heifers, such as body weight, metabolic status, heat stress, and sufficient ovarian activity, Reduced postpartum resumption of ovarian cyclicity is related to nutritional level, body condition, body weight change, and breed [9].

The gonadotropins luteinizing hormone (LH) and follicle-stimulating hormone (FSH) play important roles in ovarian function, including regulation of follicular growth, ovulation, and corpus luteum (CL) development [53]. Most studies indicate that heat stress depresses LH secretion and compromises its function. For instance, follicle tissues obtained from heat-stressed cows were shown to secrete lower levels of steroids under gonadotropin stimulation [12]. Lower concentrations of GnRH-induced LH were observed under heat stress [13].

Unlike LH, FSH secretion increases under heat stress and is associated with a larger number of follicles growing in the ovaries [14]. Rothet al. [15] also reported a pronounced decrease in plasma inhibin concentration in heat-stressed cows, which in turn caused an increase in plasma FSH concentration, known to stimulate follicle growth in the ovaries. These alterations might explain the significant rise in double ovulation and the marked rise in the calving of twins following summer insemination. Low LH surges may cause the development of suboptimal CL, which secretes low levels of progesterone. Together, altered gonadotropin secretion can depress cow fertility in the summer [54].

Reproductive performance of the cow and heifer is one of the most important factors that influence dairy sector profitability. It has been declining in dairy cows with an increasing number of days open and decreased conception rates over the last 25 years [16]. One major problem is a lack of accurate estrus detection. Most cow operations would benefit economically by reducing the number of operational days, decreasing culling rates due to non-pregnant females, and shortening their calving interval through reproductive management [17]. Successful reproductive performance based on the detection of estrous behavior requires the need to accurately detect estrous onset in the majority of cows and then inseminate 4 to 16 h later [18].

Due to intensive selection, *Bostaurus breeds* are very efficient for milk and meat production as compared to other breeds; however, they are not well adapted to tropical environments and cannot maximize their potential for production in the tropics. The main problem with Bostaurus cattle*icus (Zebu) breeds, theirbu* breeds experience a severe reduction in feed intake, growth rate, milk yield, and reproductive function in response to heat stress [19].

Effective breeding management is a crucial tool to enhance the reproductive and productive performance of dairy cattle. Knowledge of reproductive physiology, especially the differences between *Bostaurus* and *Bosindicus*, is important for the development and application of different techniques of reproductive management in cattle. However, most of the data were generated under different conditions than those observed in a tropical environment, like Brazil [20]. Furthermore, most research reports on the reproductive physiology of *Bostaurus* and *Bosindicus* females showed that *Bosindicus* had a greater number of small ovarian follicles and ovulatory follicles and a smaller corpus luteum (CL) as compared to *Bostaurus*. For instance, [21] observed a greater number of small follicles (2–5 mm) in the ovaries of Nelore (42.7 ± 5.9) than Holstein (19.7 ± 3.2) cows at the time of wave emergence and persisted throughout the estrous cycle. Nevertheless, despite ovulating smaller follicles and having smaller CL, *Bosindicus* cows or heifers have higher circulating concentrations of estradiol, progesterone, insulin and IGF-I compared to *Bostaurus females [22*].

Progesterone plays an essential role in various reproductive functions, including regulating the length of the estrous cycle, maintaining pregnancy [23], regulating embryonic growth and development (24; 25), and having been used to detect pathology of the reproductive system such as cystic ovaries [26]. Ovarian estradiol also plays an important role in establishing the timing of uterine receptivity to the developing embryo [27] and embryo survival [28]. Circulating concentrations of estradiol during the preovulatory period can influence the establishment and maintenance of pregnancy by altering the uterine environment [28, 29] and determining the appearance of behavioral estrus and subsequent ovulation [30]. In relation to circulating steroid and metabolic hormones (e.g., estradiol and progesterone), studies that have directly compared *Bostaurus* and *Bosindicus* are scarce.

Besides, heat stress during the summer substantially contributes to lower fertility and production, especially in *Bostaurus* breeds. In contrast, *Bosindicus* breeds are known for their adaptation to adverse conditions at tropical environments, especially due to their higher thermo tolerance as compared to European breeds. However, the impact of heat stress on Zebu breeds should not be ignored [31,32] and can be used as an estimative for predicting the effects of heat stress on cattle performance [33]. In relation to the heat stress influence on neuroendocrine and reproductive physiology of *Bostaurus and Bosindicus*, few studies have reported that FSH plasma concentrations are increased in cows under heat stress, probably because circulating inhibin concentrations are reduced by the impairment of follicular development [34].

Low estrus detection rate has been identified as an important factor affecting the reproductive efficiency [35]. The optimal timing of artificial insemination is signaled by estrous behavior. However, the detection of estrus in modern high milk-yield dairy cows is hindered, because the duration and intensity of estrous behavior in these cows is considerably lower than that in dairy cows of a few decades ago [35].Therefore, the objective of the present study was to (1) determine blood hormone level (Estrogen , progesterone and FSH) , (2) determine blood hormone level with body condition score and, (3) assess the behavioral estrous shown during heat period in purebred Boran and Boran×Holstein breeds of dairy cattle in at Bishoftu Agricultural Research Center, Ethiopia.

Materials and Methods

Study Area

The study was carried out from October 2019 to June 2020 at Bishoftu Agricultural Research Center (DZARC), which is located at a distance of about 45 km south east of Addis Ababa. It lies between latitude

08° 44′N &longitude 38° 58′ E at an altitude of 1900 m.a.s.l. The mean annual minimum and maximum temperatures are 8.9°C and 28.3°C, respectively, with an overall average of 19°C and an average annual rainfall of 851 mm(DZARC, 2018).

Experimental Animals Management

DZARC owns about 150 dairy animals of which 100 are Boran breeds, whereas the remaining 50 are crosses. The animals are primarily maintained for various research purpose including nutrition, diseases, reproduction and breeding. The dairy farm practices zero grazing and a free stall system, where animals are allowed to exercise in yard. There is separate laboratory for animal health and the newly established reproductive biotechnology section, and another separate laboratory for nutrition and feed analysis. Dairy Cows used in this study were managed under intensive farming system with uniform management, housing, except feeding. They were provided with feed of different mix: *tef* (*Eragrostistef)* straw and grass (*Andropogonabyssinicus*) hay as a basal diet and lactating animals were supplemented with commercially prepared concentrates based on their milk production. A total number of 20 (10 purebred Borans and 10Boran×Holstein) multiparous dairy cows were selected for this study after screening for normal reproductive anatomy through clinical and gynecological examinations. Animals with a normal estrus cycle length of 19-23 days were used for the study. Cow body condition score (BCS) was recorded at the beginning of the study. Selected cows had a mean BCS of 3 (Score 1and 2=Poor, Score 3 and 4= Medium, and Score 5= Good [36].

Sampling Method and Selection of Experimental Animals

The experimental animals were selected purposively based on parity, breed, apparently healthy, non pregnant and cyclic.

Sample Collections and Laboratory Analysis

Blood sample (10 ml) were collected on day 0 (day of standing heat), day 7, and 11 post estrous in January and Febrary2020 by vein puncture in order to measure serum Follicle Stimulating Hormone (FSH), Estradiol 17 beta (E2) and progesterone (P4) concentrations and serum were extracted and stored at -20°C at DZARC reproductive biotechnology laboratory until analysis. Samples were submitted to Ethiopian Public Health Institute (EPHI), national reference laboratory for clinical chemistry, using dry ice for analysis. Serum Estradiol concentration was measured by Elecsys Estradiol III Cobas® MODULAR ANALYTICS E170 cobas e 602analyzers having a measuring range 18.4-11010 pmol/L (5-3000 pg/mL) and serum progesterone concentration was measured by Elecsys Progesterone III Cobas® having measuring range 0.159-191 nmol/l or 0.05-60 ng/ml, serum FSH concentration were measured with ElecsysFSHCobas® having measuring range 0.100-200 mIU/ml (defined by the lower detection limit and the maximum of the master curve). Values below the lower detection limit are reported as < 0.100 mIU/ml. The concentaration of the reproductive hormones on day 0, day 7 and 11 post ovulation were evaluated .

Statistical Analysis

Both quantitative and qualitative data was entered in Microsoft excel sheet, then the data was converted to Microsoft 2002 workbook and analysed using SAS software programme version 9.00 (2002, USA). Data were summarized using descriptive statistics such as the mean ± standard error of the mean (SEM)and frequency distribution. Relationship between breeds, BCS and hormonal concentrations were analyzed by using Pearson's correlation coefficients. Independent sample t-test was undertaken to compare means between breeds of cows. Differences between the mean values were considered significant when $P < 0.05$

Results

Characteristics of Behavioral Estrus

Details of behavioral estrus characteristics were indicated in Table 1. The behavioral estrous shown during heat period were assessed and Estrus characteristics were evaluated, for 20 animals(N= 10 Boran and 10 Boran*Holstein). Results revealed that65%,55%,50%,45%,40%, 35%,30%and 25% of animals showed

standing to be mounted, redness of vaginal mucosa, vaginal mucus discharge, attempt or rejection to mount, restlessness, disoriented mounts, frequent urination, ruffled hair and bellowing, respectively. Thirty five percent of the crossbred and 30% of Boran showed quiet acceptance(standing) to be mounted. Disoriented mounts are exhibited by 25% of Boran and 10% of Crossbred dairy cattle during the estrus period, whereas attempt or rejection to mount was exhibited by30% Boran and 15% crossbred cows during the study period.

Table 5: Behavioral estrus characteristics of purebred Boran and Boran *Holstein during estrous(Between breed difference).

Estrus behavior exhibited	Total observed	Boran	cross	Boran (%)	Cross (%)	Total %
Quit acceptance(standing) to be mounted	20	6	7	30	35	65
Disoriented mounts	20	5	2	25	10	35
Chin resting	20	3		15	0	15
Ruffled hair	20	3	4	15	20	25
Redness of vaginal mucosa	20	7	4	35	20	55
Vaginal mucus discharge	20	5	5	25	25	50
Frequent urination	20	3	3	15	15	30
Sniffing (flehmann)	20	2	2	10	10	20
Restlessness	20	8		40	0	40
Attempt or rejection to mount	20	6	3	30	15	45
Soliciting with others	20	3	0	15	0	15
Bellowing	20	2	3	10	15	25

Hormone analysis

Progesterone (P4) Assay

The blood concentrations of progesterone were remarkably fluctuated according to the days and stages of estrous cycles of the cows under experimental observation. The progesterone status was significantly ($p < 0.01$) lower during day zero of estrus followed by rising of values during metestrus periods (Table 2). The greater blood concentrations of progesterone were observed throughout the length of early diestrus periods and began to decline. The minimum and maximum blood concentrations of progesterone were noted as 0.05 and1.57ng/ml on day zero 0.053 and 4.37ng/ml on day 7 and 0.161 and 3.24ng/ml day 11, respectively(Table 2).

Estrogen(E2) assay

The blood concentrations of estrogen were remarkably fluctuated according to days (0,7 and 11) and stages of estrous cycle of the cows under experimental observation. The estrogen status was significantly increased during the days of estrus (D0) and with the advancement of the days the estrogen level started to fall atmetestrus period. The lowest blood concentration of estrogen was observed throughout the length of early diestrus period and began to rise (Table 2). The average mean of Estradiol was19.38±2.46pg/ml with 5.78 pg/ml and 46.23pg/ml minimum and maximum value, respectively, on day zero (standing heat) of estrus cycle.

On day 7, the estradiol concentration was slightly decreased to 16.08±2.16pg/ml with 5 ng/ml and 41.83pg/ml minimum and maximum values, respectively. And the progesterone concentration was increased to 1.12±0.32ng/ml having 0.053ng/ml and 4.37ng/ml minimum and maximum values, respectively. Estradiol is decreased to 15.46±2.09pg/ml with 5pg/ml and 37.05pg/ml minimum and maximum observations, respectively, on day11 of the estrus cycle and the difference was statistically significant ($P<0.05$).

Follicular stimulating hormone (FSH) Assay

Serum FSH level in all cows wasobserved to be <0.100mlU/ml on day 0,7 and 11.

Table 6: Steroid hormone concentarions(Mean±SE) on days of estrus cycle,BCS and breed.

Variable	N	Estradiol(Mean±SE)	Progesterone(Mean±SE)
Days		**	***
0	20	19.38±2.46[a]	0.31±0.08[a]
7	20	16.08±2.16[b]	1.12±0.32[b]
11	20	15.46±2.09[b]	1.19±0.19[b]
BCS		**	**
Poor	9	15.4±2.18[a]	0.666±0.21[a]
Medium	33	16.21±1.49[a]	1.077±0.26[b]
Good	18	19.15±3.19[b]	1.120±0.31[b]
Breed		**	*
Boran	30	20.89±1.75[a]	1.12±0.23[a]
Boran*Holstein	30	13.05±1.63[b]	0.99±0.26[b]

N= number of observations; SE=standard error;*p<0.05;**P<0.01,***P<0.001. column with different superscript letters are significantly different.

Effect of BCS

Animals with poor body condition have on average 15.4±2.18pg/ml of estradiol(E2) concentration and 0.666±0.21ng/ml progesterone (p4)where as animal with good body condition score have a mean of 19.15±3.19pg/ml of Estradiol(E2) and 1.12±0.31ng/ml progesterone(p4), respectively, which wasstatistically significant (P<0.05).Animals with medium body condition have values 16.21±1.49pg/ml Estradiol and 1.077±0.26ng/ml progesterone(Table 2,Figure 1,Figure 2).

Pure Boran breed have high circulating Estradiol (E2)concentration than Boran*Holstein in the present studies having a mean of 20.89±1.75pg/ml and 13.05±1.63pg/ml, respectively, which is statistically significant(P<0.0001), while the progesterone profile of pure Boran and Boran *Holstein was1.12±0.23ng/ml and 0.99±0.26ng/ml, respectively,this also higher in pure Boran which wasstatistically significant (p<0.0001)(Table 2andFigure 1)

The result shows that there wasstatistically significant difference in blood estrogen concentration between breeds.Boran *Holstein breeds hadsignificantly (p < 0.01) lower (9.469 pg/ml)blood estradiol than purebred Boran.

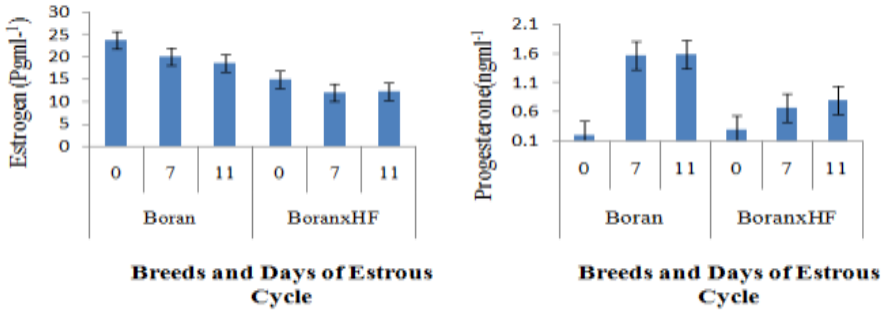

Figure 1: Patterns of Hormone (Estrogen(E2) and projesterone(P4) between breed differences during estrous cycle.

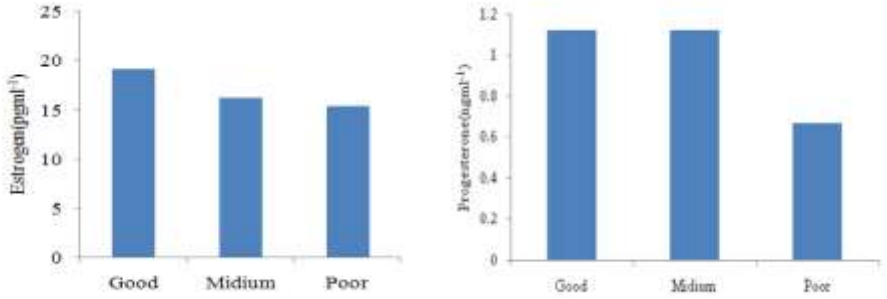

Figure 2: Patterns of Hormone (Estrogen(E2) and projesterone(P4) with BCS.

Discussions

In the current study, the concentration of Estradiol Purebred Boran(20.89±1.75pg/ml) and Boran*Holstein (13.05± 1.63pg/ml) is higher the findings of [21] who reported peak plasma estradiol concentrations in Holsteins and Nelore (7.70 ± 0.67 *vs*12.71 ± 0.98 pg/ml) and progesterone (4.06 ± 0.18 *vs*4.64 ± 0.40 ng/ml, respectively. This suggests that Boran*Holstein cows may be slower to metabolize estradiol and progesterone. But Alvarezet al. [37] reported that there were no differences in maximum circulating estradiol among Brahman, Angus, and Senepol cows (8.9 ± 1.6, 9.1 ± 1.4, and 8.7 ± 1.4 pg/ml), respectively.

The increase in estradiol concentration was associated with the decrease in progesterone concentration. In the current study, the estradiol concentrations of all cows were higher than 5.0 pg/ml, indicating that all animals have ovulated. This is in line with [38] who reported lowest serum concentrations of progesterone and highest level of serum estrogen that were noted during day zero is an indication of onset of estrus. Along with follicular growth, estradiol concentrations are associated with ovulation were >5.0 pg/ml, while concentrations < 5.0 pg/ml were not enough to induce ovulation.

The progesterone profile of purebred Boran and Boran*Holstein was1.12±0.23ng/ml and 0.99±0.26ng/ml, respectively. This result is in line with [39] who reported that under normal conditions, circulating progesterone is very low (normally, no more than 10% of the cows should have progesterone in serum greater than 1 ng/ml). Our findings are also in agreement with studies performed in Brazil which detected higher circulating progesterone in *Bosindicus* (Nelore or Gir) compared to *Bostaurus*(Angus or Holstein) cattle. [40] compared *Bosindicus* (Nelore and Gir), *Bostaurus* (Angus and Holstein) and crossbreds (Angus x Nelore and Gir x Holstein) throughout the period of an estrus synchronization protocol. Our result disagrees with the work of [41] who reports *Bosindicus* and *Bosindicus* crossbred females had lower progesterone concentration per gram of luteal tissue than in *Bostaurus* females, while[42] did not detect differences in progesterone concentration in the CL between Brahman and Angus cows (75.8 ± 11.3 and 65.9 ± 5.3 μg/ml of CL, respectively) (P > 0.10). Sartoriet al [43]also reported that possibly due to greater production, but also, a reduced metabolic clearance rate for P4 than in *B. taurus*. Moreover, among *B. taurus* breeds, lactating dairy cows have even lower circulating steroid hormones [44] due to greater metabolic clearance associated with greater dry matter intake [45]. So, the differences in value maybe attributed to breed difference as well as management practices like feeding.

In this study, progesterone level, is detectable but low (0.05-5 ng/ml) having a mean of 0.31ng/ml during D0 of the cycle in both Boran and Boran *Holstein. It began to increase during the 7th day period and reached a maximum in all cows of both breed 0.05-4.37 with mean of 1.19ng/ml on day 11th of the estrous cycle. The findings of the present study is in line with Robertson [46] who detected progesterone from plasma. In the current study, serum FSH concentrations were less than 0.100mlU/ml, and this shows that new batches of follicules did not start growing yet and they are on maturation stage.

In the present study, the mean of estradiol level from serum on D0 was 19.38± 2.46pg/ml, 16.08± 2.16 pg/ml on day 7 and 15.46±2.09pg/ml on day 11 of estrous cycle ($p < 0.05$)..This finding deviates from[47]who reported 3.09±0.02pg/ml on D0,2.94±0.01pg/ml on day 7and 2.94±0.01pg/ml on day 11with the maximal and minimal blood concentrations of estrogen that were recorded as 3.09±0.02 and 2.94±0.01 pg/ml, respectively. This difference may be arised from sample size ,environmental impact and other management practices such as feeding and lactation stage.

The mean of progesterone concentration on D0 is 0.31± 0.08ng/ml , 1.12± 0.32 ng/ml on day 7 and 1.19± 0.19ng/ml on day 11 and ,this finding is in agreement with[48] who reported 0.28±0.07,0.97±0.14 and 1.59±0.51ng/ml from follicular fluid collected from abattoir during Metestrous ,Early diestrousand late diestrous, respectively. [47]also reported 1.06±0.02ng/ml on D0,1.23±0.01ng/ml on day 7 and 1.22±0.02ng/ml on day 11,with the maximum and minimum blood concentrations of progesterone were noted as 1.26±0.01 and 1.04±0.00 ng/ml, respectively.

There is no pvevious work that relates the concentration of steroid hormone with body condition score ,but in the present work animals with poor body conditions have 15.403pg/ml and 0.666ng/ml(n=3) estradiol and progesterone, respectively whereas animals with medium body condition have 16.21pg/ml and 1.077ng/ml(n=11) estradiol and progesterone, respectively and animals with good body condition score have a serum concentration of 19.154pg/ml and 1.122ng/ml(n=6), respectively. However, Diskin et al [49] reported that inadequate nutrition can have an adverse effect on pituitary function that is the synthesis or release of gonadotropin and may also influence the response of target organs to gonadotropins or gonadal hormones and this is measured by their BCS.

The within breed differences impure bred Boran and Boran*Holsteincows in relation to Body condition score and days of estrus cycle clearly showed that there wasa mean difference in the concentration of steroid hormone and the difference was statistically significant ($p<0.05$).This differences in Boran*Holstein clearly showed that the difference in mean of Estrogen level was mainly due to days of estrus cycle which is statistically significant ($p<0.05$).This finding is in line with[44]who reported that the difference among *B. taurus* breeds could be mainly due to lactation, and lactating dairy cows have lower circulating steroid hormones due to greater metabolic clearance associated with greater dry matter intake [45].

In the present study, there was a within breed and between breed differences in estrous behavior. The purebred Boran exhibited by disoriented mounting, restlessness, attempt or rejection to mount, chin resting and soliciting with others, whereas the Boran*Holstein breeds exhibited quit acceptance (standing) to be mounted while the rest behaviors were manifested by both breeds. The within breed differences in Boran*Holstein mainly dependent on the level of milk production, since, behavioral changes during estrous was correlated to the level of estradiol within the circulation. This result is in agreement with the findingsof [35]who reported that the within breed differences may be related to lower circulating concentrations of estradiol in higher *vs* lower milk producing cows. Although there are differences between *Bostaurus* and *Bosindicus* in duration and intensity of estrus, other factors such as feed intake, body size, and steroid metabolism may exert profound effects over this behavior [50] reviewed that, behavioral signs of estrus can be observed in cattle and used for estrus detection for the purposes of artificial insemination (AI). However, these characteristics are influenced by age, milk yield, environment and hierarchy. Additionally, differences can be noted among breeds [51], and even among genetic groups (*Bosindicusvs Bostaurus*).

Conclusion and Recommendations

Understanding the reproductive physiology, especially the differences between Boran and Boran*Holstein in Ethiopia, is key for the development and application of different techniques like AI, sexed semen, MOET, ovum collection, IVF, and other current technologies in reproductive management in cattle. Purebred Boran have a higher concentration of circulating steroid hormones (estradiol and progesterone) than Boran*Holsteinbreeds. Serum estradiol concentration increases on day zero and slightly decreases in both

breeds in successive days, while progesterone concentration increases after day zero in both breeds of cattle. This lowest serum concentration of progesterone and highest level of serum estrogen that were prominent during day zero is a sign of set of estruses. The level of the steroid hormone is higher in medium- and good-body-condition cows and lower in poor-body-condition animals. There was a considerable difference in steroid hormone levels within breeds as well as between breeds. There were also differences in estrus behavior exhibited between and within breeds. Based on the above conclusion, the following recommendation was forwarded to the stakeholders:

- To increase the success of reproductive biotechnology in the dairy sector, a detailed investigation of the reproductive hormone profile must be assessed to establish the apparently normal reproductive physiology of Boran and Boran*Holstein breeds of cattle in Ethiopia.
- Quit acceptance (standing) to be mounted was a sign of heat that was exaggerated during standing heat in either of the breeds.
- Feeding cows to maintain a good BCS can improve their hormonal profile.
- Failing to detect heat is the most frequent cause of reproductive management failure. Further research should be done to identify management factors affecting the hormonal profile and causing reproduction failure.

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Assessing Ground Cover Rice Production System on Water Saving and Nitrogen Use Efficiency in Uganda

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Abstract
Rice is an important food staple and source of cash income to the majority of the populace in many parts of the world including Sub-Saharan Africa where it provides up to 21% calorie intake and accounts for 29% of the total global output of the grain crops. However, water scarcity and the continued effects of fertilisers use in rice fields makes rice both a contributor and vulnerable to climate change. A number of innovations including Ground cover rice production system (GCRPS) has demonstrated water saving and increased nitrogen use efficiencies in nitrogen fertilised rice fields, respectively although, there is a paucity of information on GCRPS in Uganda. This study was therefore conducted to assess the feasibility/viability of GCRPS for water saving and nitrogen use efficiency in Uganda. The experimental design was a randomised complete block design (RCBD) with three replications. Treatments assessed included rice planted on polyethylene mulched ground, rice planted on straw mulched ground, rice planted on bare ground and traditional flooded rice system as a control. Grain yield, straw yield, plant tissue N concentration and water use efficiency were used to assess the performance of the treatments, respectively.The mulched treatments significantly ($p<0.05$) increased grain dry matter and water use efficiency, respectively. Similarly, the straw dry-matter, % N concentration, and N uptake were increased by the mulched treatment. In fact, the poly system consistently improved nitrogen use efficiencies, which were generally comparable to the high yielding lowland rice cultivation systems reported in literature implying the GCRPS can effectively replace lowland rice ecosystems in terms of nitrogen use efficiency. Overall, the mulched GCRPS systems resulted in water saving ranging from 76% to 84% of applied water on flooded rice. Therefore, the mulched GCRPS systems can be an effective counter against N nutrient deficient soils and the declining water resource that cannot support efficient rice production in Uganda.
Keywords: Ground cover rice production systems, nitrogen use efficiency, water saving

1. Introduction
Rice (*Oryza sativa* L.) is one of the leading staple crop providing livelihoods to over 3billion people including the 1 billion poor globally (Zeiglar and Barclay, 2008, FAO, 2021). In Sub-Saharan Africa, rice provides up to 21% calorie intake as well as represents 29% of the total global output of the grain crops (FAO, 2021). According to Bua and Ojirot, (2014), the growing importance of rice as a food staple and cash crop in Uganda is attributed to the extensive urbanisation and decline in the production of finger millet, cassava and banana the major sources of carbohydrate staples in the country. The production of rice to meet the nutrition needs of an ever growing world population needs to be raised sustainably to mitigate the already present effects of rice cultivation on the environment (Fukagawa and Ziska, 2019). Nevertheless, traditional rice production in flooded and upland non irrigated fields has been preferred in most rice growing regions due to the farmers knowledge of practices used for rice production (GRiSP, 2013). The traditional rice growing ecosystems have faced a myriad of challenges overtime such as frequent droughts, high fertilizer prices, labor scarcity, weeds and pests infestations (Shaobing, Qiyuan, and Yingbin, 2009). This has highlighted the need for more research to curtail these problems. Furthermore, water scarcity is

increasingly becoming a serious threat to agricultural production affecting crops such as rice with higher water use efficiency (Mancosu, Snyder, Kyriakakis, and Spano, 2015). According to IRRI (2009), water scarcity affects more than 23 million hectares of rain-fed rice production areas in south and south-east Asia whereas recurring drought affects nearly 80% of the potential 20 million hectares of rain-fed lowland rice in Africa. Moreover, the contamination of the environment, water bodies and degraded upland areas as well as varieties less adapted to climate change further exacerbates the problem (Ali *et al.*, 2020; Gu and Yang, 2022). Yet, rice growing in both the lowland and upland ecosystems continues to dominate the cropping systems (GRiSP, 2013).

Therefore, one of the options being explored for water saving and improved nitrogen use efficiency in rice production is the ground cover rice production systems (GCRPS) (Qu *et al.*, 2012). According to Lv *et al.* (2019) the direct seeded GCRPS is reported to maximise root development as a result of either application of plastic or straw mulch to conserve soil water. Moreover, water savings of between 61 - 81% and a reduction in total input water of 37 - 47% compared to the flooded rice production system were associated with the GCRPS usage in rice production in countries such as China (Jin *et al.*, 2016). The GCRPS has also been used on a wide range of soil types with consistent improvements in economic yields for rice in China (Tao *et al.*, 2006; Tao *et al.*, 2014*).* However, in Uganda, information on GCRPS is scanty and limited. Therefore, the objective of this study was to assess the feasibility/viability of GCRPS for water saving and nitrogen use efficiency in rice production systems.

2. Materials and Methods

2.1. Study Area

The study was conducted in Lira district, northern Uganda during the period October 2021 and April 2022, respectively. Lira district is bordered by Pader district in the north, Alebtong district to the east, Dokolo district in the south east, Kole and Apac districts to the west and south west, respectively. Lira district consist of 5 sub counties but the study was conducted in Itek sub-county, Ayel parish in a farmers' field (Lira District Local Government, 2015). Lira district was chosen because it is one of the major rice cultivation districts in northern Uganda (Galipac and Elepu, 2014; Akongo *et al*., 2017).The site lies on the coordinates $02^0.11'55.4^0$N $033^0 05'21.3^0$E. The annual rainfall distribution over the two seasons (March-June) and (August-December) are 644 mm and 605mm, respectively (AQUASTAT). The annual temperature ranges between 30^0 and 15^0 C, respectively. The area is characterised by the moist and sub-humid climatic conditions (Kumakech *et al.*, 2014). The soils are sandy loamy in texture (Otim *et al.*, 2015).

Weather Data

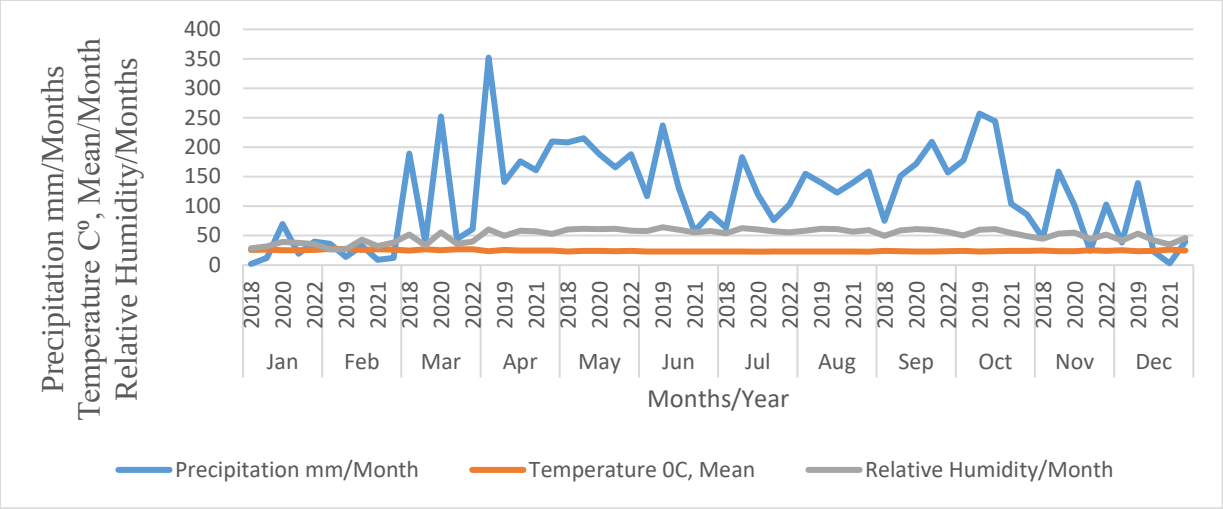

Figure 1: Precipitation (mm), Temperature (0C), Mean and Relative Humidity per month for five years 2018 - 2022 retrieved from AQUASTAT information tool for Ngetta ZARDI meteorology station, Lira.

2.2. Soil characteristics

Soil sampling from the study site was accomplished using the zig-zag method whereby soil samples were randomly taken from cores in the top 0–20 cm of the field, bulked into a plastic bucket, and thoroughly mixed into a composite sample of one kilogramme which was taken to Kyambogo university for laboratory analysis. (Imakumbili *et al.,* 2022). Soil physical and chemical properties were determined as shown in Table 1.

Table 7: Physical and chemical analyses of soil samples from farmer's field in Itek-Okile irrigation scheme, Lira district, 2021/2022

Soil Characteristics*	**First season 2021**	**Second season 2022**	**Method of analysis**
pH	5.48	6.03	Measured in water suspension
SOM (%)	5.5	7.5	Weight loss on ignition at 400°C (Faithfull, 2002)
Total N (%)	0.08	0.03	Kjedhal method (Seki, 1990).
Available P (%)	0.15	0.22	P extracted using Bray 1 solution followed by atomic absorption spectroscopy
Available K(%)	2.89	2.04	K ammonium ethanoate reagent followed by flame spectroscopy
Soil texture	Clay	Clay	Physical investigation (Moorberg and Crouse, 2021).

*The critical values for the chemical soil characteristics are pH=5.6, Soil Organic Matter (OM)%=3, Total N (%)=0.20, Phosphorus P(%)=0.0015, and Available K (%)=0.0133.

2.3. Experimental design

The experimental design was a Randomized Complete Block Design (RCBD) with three replications for each of the two seasons. The treatments assessed on the rice variety PR-107 included i) T1-Traditional flooded rice plots, ii) GCRPS straw-Straw mulched rice soil with rice plants + Urea, iii) GCRPS poly-Polyethylene mulched soil with rice plants+ Urea, vi) GCRPS bare-Bare soil planted with rice. Each

experimental plot measured 3metres x 2.5 metres ($7.5M^2$). For the flooded rice plots, a sunken basin was used as the plot area meanwhile for the raised bed plots, a total of three strips with dimensions 1metre x 2.5metres and alley of 0.5m between strips and one (1) metre between the blocks were used. Each strip was enclosed with a shallow furrow 0.15m deep and 0.20m wide while the traditional paddy rice treatment was surrounded by polyethylene sheets buried 0.60m deep to prevent horizontal movement of water between treatments. The total area covered by the experiment was 183.5 m^2 (Tao *et a*l., 2014).

2.4. Field management activities

Ploughing was carried out using ox-plough before the land was harrowed and no puddling was applied. The experimental plots were marked out ready for construction of levees. The whole experiment was enclosed with a main levee leaving an inlet and outlet. Prior to planting the whole experimental area was drained and four rice seeds were planted direct into the wet soil for each treatment at a spacing of 0.30 m x 0.15m, respectively. Fertilizer was applied as a basal application two weeks after germination for the GCRPS at a rate of 120 Kg/ha (Tao *et al.,* 2006). For each of the treatments, irrigation water was applied up to the 0.1m mark which was measured by considering the floor of treatment and height of water layer along the levee of the treatment for the flooded rice treatment meanwhile for the GCRPS the furrows enclosing the plot were considered and furrow walls were used to measure the 0.1m layer of water. The amount of water was computed from the dimensions of the treatment plot or furrows after-which the total amount was then derived. In the first season, the flooded rice received seven irrigation applications while the GCRPS received 4 irrigation applications. However, during the second season, flooded rice received 5 irrigation applications and GCRPS 2 irrigation applications, respectively. The computation of amount of irrigation water applied to the field was based on computation considering the dimensions of the treatment plot holding water and the furrows.The straw mulch was applied to a depth of 0.15m. Over the season, straw was added to maintain the depth of mulching. A polyethylene sheet was used to mulch the GCRPS poly treatment. The polyethylene sheet mulch had round holes (measuring 5 centimeters in diameter) punched using a cork borer. The holes were to allow the dibbling of rice seeds and tillers to grow easily from the mother plant (Lv *et al.,* 2019). Rice seeds were planted direct into the raised beds and lowland treatments. Fourteen (14) days after germination, light flooding was applied for the traditional lowland rice treatment (Marasini *et al*., 2016).

2.5 Data Collection

Grain yield, straw yield, tissue N concentration were used as the indicator parameters for the performance for the different treatments. The matured rice was harvested by cutting the rice stalks at ground level from two middle rows of each plot which was equivalent to one square metre of the treatment plot. The rice stalks with the grain attached were manually threshed and winnowed to remove all foreign material, after which they were placed in a polythene bag for storage. The rice grain was then sunshine dried to 14% moisture content. The rice stalks were also tied in a single bundle for each treatment after which they were placed on wooden racks in a room with free flowing air to dry. Harvesting for the first and second season was done on 25/1/2022 and 20/8/2022, respectively. The samples were then transported to Kyambogo soil science laboratory were measurements were done using a laboratory beam balance with a 500 grams limit. The recorded weights for each treatment in kilogrammes per square meter were then converted to tons per hectare.

Separate harvests of the grain and straw samples were made earlier than other samples and where washed with water by placement of the samples on a mesh and allowing running water to pour over them for five minutes so as to remove any dust and soil particles. The samples from the different plots were then left to dry under a shade with adequate flow of air inside a room on wooden racks (Faithfull, 2002). The samples where then transported to Kyambogo University, Soil Science laboratory where they were processed and

plant samples were analysed for N using the Kjedhal method (Seki, 1990). The calculation of water use efficiency as a ratio of grain output in kilogrammes per hectare divided by the total applied irrigation water to the field was used to determine the irrigation water use efficiency or IWUE (Qin *et al*., 2006).
Nitrogen utilization efficiencies were computed as follows:

I) **Agronomic Efficiency (AEY)** is the amount of grain produced for each applied unit of nitrogen. It was calculated by using the following equation:

$$\text{AEY } (Kg\ Kg^{-1}) = \frac{\text{fertilized treatment yield in kg} - \text{unfertilized treatment yield in kg}}{\text{Amount of nitrogen applied in kg}} \quad (1)$$

II) **Utilisation Efficiency (UE)** is the product of apparent recovery efficiency and physiological efficiency

$$\text{UEY } (Kg\ Kg^{-1}) = \text{PEY.AREY} \quad (2)$$

III) **Physiological Efficiency (PEY**) is the grain plus straw yield achieved per unit of nitrogen absorbed. It was calculated using. the following equation;

$$\text{PEY}(Kg\ Kg^{-1}) = \frac{\text{A(kg)} - \text{B(kg)}}{\text{AN(kg)} - \text{BN(kg)}} \quad (3)$$

where AN= Grain and straw N uptake of fertilized plot
A= biological yield (grain plus straw) of fertilized plot
B= Biological yield of unfertilized plot and
BN= Biological yield (grain plus straw) of unfertilized plot.

IV) **Agro physiological Efficiency (APE**) is the economic output achieved per unit of nitrogen absorbed. It was calculated using the following equation:

$$\text{APEY } (Kg\ Kg^{-1}) = \frac{\text{A(kg)} - \text{B(kg)}}{\text{NA(kg)} - \text{NB(kg)}} \quad (4)$$

Where NA= Grain plus straw N uptake of fertilized treatment
NB=Grain plus straw N uptake of unfertilized treatment
A= grain yield of fertilized treatment and
B= grain yield of unfertilized treatment

V) **Apparent Recovery Efficiency (AREY)** refers to the amount of nitrogen absorbed for each applied unit of nitrogen. It was calculated using the following equation:

$$\text{AREY (\%)} = \left(\frac{\text{A} - \text{B}}{\text{amount of nitrogen applied}}\right) * 100 \quad (5)$$

Where A=Grain and straw nitrogen uptake of the treatment and
B= Grain and straw nitrogen uptake of the unfertilized treatment (Baligar and Fageria, 2015).

2.5. Data Analysis

The significance of the treatments was determined using Analysis of Variance (ANOVA) of the Genstat computer programme version 14. Where means were significantly different, they were seperated using the Fishers LSD at 5% probability levels (Gomez and Gomez, 1984).

3. Results

3.1. Water requirement

In both years Water Use Efficiency significantly differed (P<0.05) among the rice cultivation systems. However, during the first season, the highest WUE ($\boldsymbol{KgM^{-3}}$) was recorded from GCRPS straw. In contrast, during the second season, the highest WUE ($\boldsymbol{KgM^{-3}}$) was recorded from GCRPS poly treated plots (Table 2). The mulched GCRPS systems recorded consistently higher increase in WUE as opposed to the flooded rice plots. In fact, there was a 76% and 84% decrease in water applied to the GCRPS systems in 2021 and 2022, respectively.

Table 2: Water Use and saving of the Ground Cover Rice production System relative to the flooded rice, Okile-Itek, Lira district 2021/2022

Treatment	Grain Yield ($Kgha^{-1}$)	Water applied (Kg/ha)	WUE (KgM^{-3})	Water saving (Kg/ha)	% Increment WUE relative to flooded rice
First season October 2021					
Flooded rice	3640	6933	0.59A	-	-
GCRPS straw	4370	1653	6.21B	5280	952.5
GCRPS bare	3560	1653	3.41C	5280	477.9
GCRPS Poly	5050	1653	6.18D	5280	947.4
Second season April 2022					
Flooded rice	2950	5000	0.52A	-	-
GCRPS straw	4960	800	2.60B	4200	400
GCRPS bare	2730	800	2.10C	4200	303.8
GCRPS Poly	4940	800	3.00D	4200	476.9

At the 5% probability level, there are no detectable differences between values with similar capital letters.

3.2. Dry matter yield and percent N content at harvest

During the first season, significantly higher grain dry matter yield was recorded from GCRPS poly compared to the flooded rice system and GCRPS bare system, respectively. Conversely, during the second season, there was no significant different (P>0.05) in grain dry matter yield between the mulched and GCRPS systems. Overall, during both seasons, GCRPS bare recorded the lowest dry matter compared to the flooded rice treatment. A similar trend was followed by the straw dry matter yield. In fact, the mulched GCRPS systems recorded higher straw dry matter yield compared to the flooded rice and GCRPS bare, respectively (Table 3). The concentrations of N in the straw plant material during the first and second season ranged from 0.42 - 0.59% and 0.39 - 0.82, respectively. However, GCRPS poly and flooded rice consistently recorded higher %N concentration in the grain at harvest although the %N concentration in straw was low during both seasons except for GCRPS bare, GCRPS straw and flooded rice systems during the second season.

Table 3: Grain and straw dry matter yield, and % nitrogen concentration of grain and straw of rice grown under ground cover rice production systems at Okile-itek, Lira district, 2021/2022

Treatment	**Grain Dry Matter ($Kgha^{-1}$)**	**Straw Dry matter ($Kgha^{-1}$)**	**At harvest**	**%N**
First season October 2021				
			Grain	Straw
GCRPS Poly	5050A	5820A	0.60	0.48
GCRPS straw	4370BA	5310A	0.60	0.59
Flooded rice	3640C	4560A	0.74	0.42
GCRPS bare	3560D	4170A	0.59	0.50

Second season April 2022				
GCRPS Poly	4940A	6980A	0.63	0.39
GCRPS straw	4960BA	6710A	0.32	0.82
Flooded rice	2950C	4080A	0.59	0.69
GCRPS bare	2730D	4460A	0.42	0.60

At the 5% probability level, there are no discernible differences between values with similar upper case letters.

3.3. Nitrogen uptake and Nitrogen Harvest indices

During both the first and second season, a higher total N Uptake was recorded from the mulched GCRPS system followed by flooded rice system and GCRPS bare system, respectively. Interestingly, the GCRPS poly had the highest constant total N uptake of all the system during the two seasons. However, in comparison to GCRPS poly, GCRPS straw showed a total N uptake that was 17.7% higher. In relation to other systems, the lowest nitrogen uptake from the grain and straw was recorded from the GCRPS bare during both seasons. Conversely, the Nitrogen Harvest index (NHI) varied from 0.29 to 0.58 for all treatments during the two seasons but was constant at 0.52 for GCRPS poly (Table 4).

Table 4: Nitrogen Uptake and Nitrogen Harvest indices of lowland rice grown under different rice cultivation systems at Okile-itek, Lira district, 2021/2022

Treatment	Grain N ($Kgha^{-1}$)	Straw N ($Kgha^{-1}$)	Total N ($Kgha^{-1}$)	NHI
First season October 2021				
Flooded rice	26.94	19.15	46.09	0.58
GCRPS straw	26.22	31.33	57.55	0.46
GCRPS bare	21	20.85	41.85	0.50
GCRPS Poly	30.3	27.94	58.24	0.52
Second season April 2022				
Flooded rice	17.40	40.8	58.20	0.29
GCRPS straw	15.87	55.02	70.89	0.22
GCRPS bare	11.47	26.76	38.23	0.30
GCRPS Poly	31.12	27.22	58.34	0.53

3.4 Nitrogen Use efficiencies

The five most critical nitrogen use efficiencies for agricultural plants determined is presented in Table 5. All the nitrogen use efficiencies increased during the second season for both GCRPS treatments when nitrogen was applied at the rate of 120 kg per hectare. However, in 2022 Physiological efficiency PE decreased by 73.7% and Apparent Recovery Efficiency ARE increased by 108% in the GCRPS straw system. Nitrogen Utilization efficiency NUE for the GCRPS poly in both years was high while NUE decreased by 45.3% in GCRPS straw in 2022 compared to 2021. In general, across both years the average of means for GCRPS poly system, for nitrogen use efficiencies were calculated as shown below; Agronomic efficiency was 15.42 kilograms of grain produced for every kilogram of nitrogen applied, physiological efficiency was 213.3 kilograms of biological yield (grain and straw) for every kilogram of nitrogen accumulated, agro-physiological efficiency was 100.7 kilograms of grain produced for every kilogram of nitrogen stored in the grain and straw, apparent recovery efficiency was 15.2%, and nitrogen use efficiency produced 32.77 kilograms of grain produced for every kilogram of nitrogen used.

Table 5: Different nitrogen use efficiencies assessed under two ground cover rice production systems at Okile-itek, Lira district, 2012/2022

First season October 2021					
Different Nitrogen Use Efficiencies*					
Treatment	AEY ($KgKg^{-1}$)	PEY ($KgKg^{-1}$)	APEY ($KgKg^{-1}$)	AREY (%)	UEY ($KgKg^{-1}$)
GCRPS Poly	12.42	191.5	90.90	13.65	26.15
GCRPS straw	6.75	124.2	51.59	13.08	16.24
Second season April 2022					
GCRPS Poly	18.42	235.21	110.5	16.75	39.39
GCRPS straw	18.58	32.66	68.27	27.21	8.88

**APEY is for agro-physiological efficiency, PEY stands for agronomic efficiency, AREY stands for apparent recovery efficiency and UEY stands for utilisation efficiency.*

4. Discussion

This study has shown the possibility and potential for attaining high water use efficiency and thus saving on water used in rice fields, while improving nitrogen use efficiencies of a lowland rice variety grown in conditions similar to upland ecosystems. The GCRPS poly had the highest yield among the rice cultivation systems. Similar results were reported by Guo *et al.,* (2020) and Tao *et al.,* (2006), in which the rice grain yield for the GCRPS poly system was greater than or equal to that for flooded rice. However, most earlier research on water use had placed greater emphasis on describing how GCRPS alters grain production compared to the nitrogen fertilised flooded rice system. The popularity of the GCRPS system is also hinged on it's effective and efficient water saving ability. Therefore, this results has shown that there was a decrease in water applied to the GCRPS system by 76% and 84% during the two seasons, respectively. Meanwhile, the percentage increments in WUE relative to flooded rice system ranged from 303.8 to 947%, respectively. This therefore implies that water applied in mulched GCRPS was utilised by rice plants to produce more grain. According to Shizhang *et al.* (1994), this was achieved by reducing rice water consumption which comprised of transpiration, evaporation, and field percolation. This perhaps mean that most of the water was used for forming and increasing grain yield. Subsequent studies have also demonstrated the ability of the mulched GCRPS systems to conserve more water and increase the root growth in the soil to provide for more water needs meant for rice plant grain formation (Zhanga *et al.,* 2017). Therefore, GCRPS would be a more effective approach to growing lowland rice varieties in upland environments due to less irrigation water being used.

The mulched GCRPS registered the highest grain and straw dry matter over the GCRPS bare and flooded rice. This could have been due to the response of the lowland rice under mulching to increased water availability and nitrogen in the soil, moreover rice is a nitrogen responsive crop (Rahman *et al*., 2007). The findings therefore demonstrated that the addition of mulch and nitrogen had a favorable impact on the grain and straw yields during both season. Bhuiyana *et al*. (2017) reported similar results as nitrogen fertiliser application increased rice dry matter production. Similarly, He *et al.,* (2013) had also shown that plastic mulching and irrigation improved dry matter production in both upland grown lowland rice, respectively.

The %N concentration varied between the years an indication that water availability in the soil and nitrogen fertilisation could have caused these effects. Ye *et al*. (2014) noted that the concentration of Nitrogen in rice plants exposed to different water regimes and nitrogen fertilisation, remarkably influenced plant tissue N concentration at harvest. This indicates that rice cultivation systems influence rice plant tissue concentrations.

Mulching improved the nitrogen uptake and nitrogen harvest index in the mulched systems. This suggests mulching increased the amount of water that was available to the rice plants, which boosted the plant's ability to absorb nitrogen from the soil due to the rice plant roots' ability to procure N in soil solution. Similarly, studies have shown mulching to improve the soil temperatures which enhances root growth (Lv *et al.,* 2019). These results were similar to the findings of Li *et al.,* (2018), who also reported that the mulched GCRPS systems benefited at the start of the growth season, from a combination of greater soil temperature and less nitrogen loss due to reduction in non-physiological water consumption leading to an increase in the quantity of nitrogen that was available to plants. Certainly this led to more nitrogen being absorbed by the plants which resulted in high N in the plant tissues. Therefore, the results indicate mulched GCRPS systems improved nitrogen uptake in the rice plant, particularly in the grain.

The nitrogen use efficiencies for the GCRPS poly system were high in both seasons.The agronomic efficiency of lowland rice has been shown to vary from 6 to 25 Kilogrammes grain per Kilogramme N in the tropics (Yoshida, 1981; Cassman *et al.,* 1996; Haefele *et al.,* 2008), results are within those limits. Tao *et al*. (2015) also reported findings which were within those limits for the mulched GCRPS as the AEY ranged from 10 to 11.6 Kilograms of grains for every Kilogram of nitrogen. In comparison to the results findings for AEY were lower, this could have been due to differences in the soil textures between the study sites (Tao *et al.,* 2015). The Physiological efficiency is determined based on biomass which includes straw and grain produced for every Kilogram of nitrogen accumulated hence the PEY showed high values in both seasons compared to the Agro-physiological efficiency. The Apparent recovery efficiency AREY fluctuated between 13.08 - 27.21%. Accordingly, Dass *et al.* (2015) reported an ARE of 44%, other studies have also shown ARE of 9 - 23% for rice (Haefele *et al.,* 2008) but in the tropics it usually ranges between 30% and 50% (Prasad and De Datta, 1979), Therefore, the results are approximately within the boundaries. However, the low N recovery from the upland soil conditions of applied N could be attributed to natural processes such as volatilization or leaching. Ju *et al.* (2009) demonstrated that in lowland conditions ammonium ion is predominantly absorbed by the rice plants directly, also in aerobic conditions, the nitrification of ammonium ions to nitrate ions occurs this exposes the accumulated dominant Nitrite ion in soil to increased instances of loss through leaching and volatilization. Khatun *et al*. (2015) reported a utilization efficiency for lowland rice that ranged between 9 - 37 Kilograms grain for every Kilogram nitrogen, however, the UEY for production of grain in the tropics is about 50 kilogrammes of grain produced for every kilogram of nitrogen (Yoshida, 1981). The results are approximately within the limits. Consequently, GCRPS demonstrates it is potential to replace the lowlnad rice cultivation system as a more efficient means to convert nitrogen consistently into economic yields for rice grain in Uganda at a nitrogen fertiliser rate of 120 KgN/Ha.

5. Conclusions

The findings suggest that the mulched GCRPS substantially ($p<0.05$) increased grain dry matter and water use efficiency. The straw dry-matter,N uptake and %N concentration were also improved in the GCRPS poly over than the other cultivation systems. The GCRPS poly system consistently improved physiological, agrophysiological, apparent recovery, and utilisation efficiency. The nitrogen use efficiencies were generally comparable to the high yielding lowland rice cultivation systems in reviewed literature, which implied the GCRPS can effectively replace lowland rice ecosystems in terms of improving nitrogen use efficiency. The mulched GCRPS systems also caused water saving of between 76% to 84% of applied water on flooded rice. Therefore the mulched GCRPS systems can be an effective counter against low dry matter yields in N nutrient deficient soils, nitrogen contamination of the environment and a declining water resource that cannot support efficient rice production.

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Phenotypic Characterization of Cassava (*Manihot esculenta* Crantz) Germplasm in Kenya

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Abstract
Cassava (*Manihot esculenta)* is an important cash crop for many small scale farmers in Kenya. The cassava genetic resources in Kenya are often underestimated due to improper characterization of the germplasm available. The objective of this study was to characterize popular cassava landraces and improved varieties grown by small-scale farmers based on their phenotypic traits. The materials were collected from seven major cassava growing counties in Kenya. The survey collected 131 cassava genotypes. These were planted at two experimental sites. Both quantitative and qualitative phenotypic traits data was collected at 3, 6, 9 and 12 months after planting. The data was subjected to Multivariate analysis and dendogram developed at $p < 0.05$. Analyzed phenotypic traits categorized the genotypes into four cluster groups. Cluster 1, 2, 3, and 4 had 72.5%, 16.0%, 3.1% and 8.4% genotypes respectively. Out of the 25 phenotypic characters assessed, a total of 11 principal components (PCs) trait sets accounted for 71.58%cumulative genetic variation at $p<0.05$.A follow up study on genetic characterization should be done to show the correlation between genetic and phenotypic characterization. Results from this study will assist farmers and breeders to optimize utilization of cassava germplasm for food security.
Keywords: Phenotypic characterization, Cassava, cluster analysis, Principal components, Kenya

Introduction
Cassava (*Manihot esculenta* Crantz) is mainly cultivated in tropical countries, particularly in sub-Saharan Africa, South America and Asia as an important staple food (FAOSTAT, 2020). Worldwide, cassava production is estimated to be 277 million tons on approximately 24.5 million hectares and provides food for more than 800 million people (FAOSTAT, 2020). According to Leon-Pacheco *et al.*, (2020), cassava crop rank third in terms of carbohydrate food source in the tropics after rice and maize and provides more than 60% of the daily calorific needs of the populations in tropical Africa and Central America. In Africa, over 90% of cassava produced is consumed as human food with only 6% devoted to livestock feed (Okbenin *et al.* 2013; Adu *et al*., 2018). Cassava production in Africa is estimated at 160 million tons on 18 million hectares and Nigeria being the largest producer country of cassava (FAO, 2018). According to FAO (2018), Eastern Africa production is estimated at 30 million tons on 3 million hectares. Tanzania leads in production with 5 million tons, followed by Uganda, Burundi, Rwanda, Kenya and South Sudan. According to the Kenya Ministry of Agriculture Annual Report (MOA, 2022), the country currently produces one million metric tons annually and has the potential to produce 3 million tons. Cassava production in Kenya takes

place mainly in the low altitude areas of the coastal region, mid-altitude zones in central and eastern regions and in Nyanza and western regions (Mware *et al*., 2009). The area under cassava in western, Nyanza and central/eastern regions is estimated at 49,000, 34,000 and 19,000 ha respectively, representing nearly all the country's total area under this crop.

In Kenya, some of the challenges in domestication of cassava are attributed to the need of possession of a great part of the biological diversity and traditional knowledge associated with this species (Masinde *et al*, 2018). Farmers partly use morphological characteristics and other attributes to distinguish, manage and conserve varieties they grow on farm. An obstacle to the reliable identification of cultivars is the existence of considerable linguistic polymorphism. Each region or farmer community has its own unique series of names for different or same cultivars with specific meaning. This informal naming of varieties, however, can lead to overestimation or underestimation of crop diversity because the same variety can take different names between regions and communities. Smallholder farmers are important guardians of crop genetic diversity beyond the centers of origin. Traditionally, farming communities are known to maintain knowledge of this genetic diversity through vernacular names. Since cassava was introduced in Kenya, farmers have named the varieties according to the source of planting material, the distinguishing phenotypic characteristics or the cyanide content. Thus the same cassava genotype may be given different names in different areas depending on the farmer's perception. For example, the variety *Adhiambo lera* (clean lady) in one area, *Karembo* (beautiful*)*, in another area, or *Kasukali* (sweet) elsewhere. In another scenario Kenya experienced a severe outbreak of Cassava Mosaic Disease (CMD) in the 1980's and 1990's. The Kenya Agricultural Livestock Research Organization (KALRO) in collaboration with the International Institute for Tropical Agriculture (IITA) introduced Tropical Manioc Selection (TMS) cassava varieties. After testing for agronomic adaptability, suitable varieties were released to farmers for growing. The varieties were released to farmers with their original coded numbers such as MM96/4466, MH95/0183, MM96/0067, SS4, KME-4. For failure to memorize these codes, farmers named all these genotypes, "*Agriculture*".

Germplasm characterization is an important aspect of cassava breeding and conservation. It involves the evaluation and documentation of genetic diversity of a collection of cassava genotypes, which is important for the development of improved varieties and for maintaining genetic diversity of crops (KEPHIS, 2016; Tumuhimbise *et al*, 2016). Some of the important parameters that are used for characterization of germplasm in cassava include: Phenotypic/morphological, biochemical, molecular, agronomic, and phonological (Okogbenin *et al*. 2013). Phenotypic identification of plants is commonly based on the morphological traits assessed and recorded in the field (Fukuda *et al.* 2010). Different cultivars have been distinguished by phenotypic characteristics, such as color and shape, branching habit, plant height, color of stem and petiole, root shape and root skin color, time of maturity, yield and the cyanogenic glycosides content in the roots (Fukuda *et al*., 2010; Saravanan, 2016).

The objective of this study was to characterize the existing popular local cassava varieties grown by small scale farmers in Kenya.

Materials and Methods
Map of the Study Area

Figure 1: Map of Kenya, where the research was undertaken.
Source: Adopted from Google map (10/6/2023)

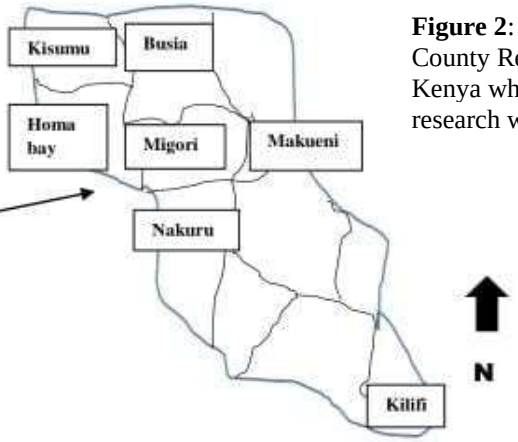

Figure 2: Sampled County Regions in Kenya where the research was done.

Source: Adopted from Google (10/6/2023)

Cassava genotypes collection and multiplication

Surveys were carried out in 2018 – 2019 main rain season and short rain season in the major cassava growing regions in Kenya. The objective was to collect the popular cassava improved varieties and landraces grown by farmers. The regions were represented by the following counties:Coastal region (Kilifi), Eastern and Central (Makueni and Nakuru), Western (Busia), Nyanza (Migori, Homabay and Kisumu). During collection, a single stem from one plantwas collected to represent the cassava accession. The local name of the variety was obtained from the farmer and also the source of the planting materials. The stem was cut into pieces length 15 cm and placed in a collection bag. The samples collected were planted in single rows for multiplication at the Rongo University farm located in Migori County. Each single row had 5 – 10 plants. The spacing was 1m between rows and 1 m within rows.

Experimental design and plot layout

The accessions were harvested from the multiplication block in January-February, 2021. Two experimental sites were established: Rongo University (-0.826279^{o}, 34.614186^{o}) and Mawego Technical Training Institute (-0.39652^{o}, 34.77068^{o}). The plants harvested from each accession were cut into pieces with each having 4-5 nodes. Each accession was planted three rows, with each row planted 5 plants. The spacing was 1m between rows and 1m between plants. Normal agronomic practices were carried out during the experimental period. No fertilizer or pesticides were applied on the crop.

Data Collection

Phenotypic data was collected on the plants in the middle row of each accession. Phenotypic characterization was done using the selected morphological and agronomic descriptors for the characterization of cassava as described by (Fukuda *et al*., 2010) (Table 1). The observations were made at 3, 6, 9, and 12 months after planting (MAP).

Table 1: Selected qualitative and quantitative phenotypic descriptors for the characterization of 131 cassava genotypes

SN	Trait descriptor	Score code	Sampling time
1	Colour of apical leaves	3 = light green; 5 = dark green; 7 = purplish green; 9 = purple	3 MAP
2	Shape of central leaflet	1 = ovoid; 2 = elliptical-lanceolate; 3 = obovate-lanceolate; 4 = oblong-lanceolate; 5 = lanceolate; 6 = straight or linear; 7 = pandurate; 8 = linear-piramidal; 9 = linear-pandurate; 10 = linear-hostalobalate	6 MAP
3	Petiole colour	1 = yellowish-green; 2 = green; 3 = reddish-green; 5 = greenish-red; 7 = red; 9 = purple	
4	Leaf colour	3 = light green; 5 = dark green; 7 = purple green; 9 = purple	
5	Number of leaf lobes	3 = three lobes; 5 = five lobes; 7 = seven lobes; 9 = nine lobes; 11 = eleven lobes	
6	Length of leaf lobe	Measurement of two middle leaf lobes from three middle plants	
7	Width of leaf lobe	Measure the width of the widest part of the same lobes in SN 6 above	
8	Ratio of lobe length to lobe width of central lobe	Calculation	
9	Lobe margins	3 = smooth; 7 = winding	
10	Petiole length	Measure two leaves per plant	
11	Colour of leaf vein	3 = green; 5 = reddish-green in less than half of the lobe; 7 = reddish-green in more than half of the lobe; 9 = all red	
12	Orientation of petiole	1 = inclined upwards; 3 = horizontal; 5 = inclined downwards; 7 = irregular	
13	Flowering	0 = absent; 1 = present	
14	Pollen	0 = absent; 1 = present	
15	Prominence of foliar scars	3 = semi-prominent; 5 = prominent	9 MAP
16	Colour of stem apex	1 = orange; 2 = light green; 3 = dark green	
17	Colour of stem epidermis	1 = cream; 2 = light brown; 3 = dark brown; 4 = orange	
18	Colour of stem exterior	3 = orange; 4 = greeny-yellowish; 5 = golden; 6 = light brown; 7 = silver; 8 = grey; 9 = dark brown	
19	Distance between leaf scars	3 = short < (8 cm); 5 = medium (8-15 cm); 7 = long > (15 cm)	
20	Growth habit of stem	1 = straight; 2 = zig-zag	
21	Colour of end branches of adult plant	3 = green; 5 = green-purple; 7 = purple	
22	Extent of root peduncle	0 = sessile; 3 = pedunculate; 5 = mixed	12 MAP (at harvest)
23	Root shape	1 = conical; 2 = conical-cylindrical; 3 = cylindrical; 4 = irregular	
24	External colour of storage root	1 = white or cream; 2 = yellow; 3 = light brown; 4 = dark brown	
25	Colour of root pulp (parenchyma)	1 = white; 2 = cream; 3 = yellow; 4 = orange; 5 = pink	
26	Colour of root cortex	1 = white or cream; 2 = yellow; 3 = pink; 4 = purple	
27	Cortex: ease of peeling	1 = easy; 2 = difficult	
28	Texture of root epidermis	3 = smooth; 5 = intermediate; 7 = rough	
29	Root taste	1 = sweet; 2 = intermediate; 3 = bitter	
30	Cortex thickness	1 = thin; 2 = intermediate; 3 = thick	

Data Analysis

The genetic variation among the studied genotypes for agro-morphological traits was explored using multivariate analysis technique (Karim *et al*. 2020). Multivariate analysis of the 131 data matrix comprising of principal component analysis (PCA) processed using IBM SPSS statistics software version

25. In the PCA, Eigenvalues and load coefficient values were generated from the data set. The relevance of trait contribution to the variation accounted by each principal component was based on the absolute eigenvector arbitrary cutoff value of 0.30 (Richman, 1988). Structure of morphological changeability was visualized using ascending hierarchical clustering (AHC) based on data and Ward's Method to plot a dendrogram (Karim *et al*., 2020). The principal component analysis and correlation matrices were used to determine the relationships among the traits.

Results

Principal component analysis of phenotypic characters

The eigenvalues and percentage variations of the principal component analysis are presented in Table 2. Eigenvalues are the special set of scalar values that is associated with the set of linear equations most probably in the matrix equations. Eleven principal components that accounted for 71.58% of the total variation among the genotypes were identified. The first PC axis with eigenvalue of 3.27 accounted for 13.07% of the total variation whereas the second, third, fourth and the fifth PC axes with eigenvalues of 2.39, 2.04, 1.74 and 1.54 accounted for9.55%, 8.15%, 6.97% and 6.15% of the total variation, respectively. The sixth, seventh, eighth, ninth, tenth and eleventh PC axes with eigenvalues of 1.41, 1.27, 1.16, 1.11, 0.99 and 0.98 accounted for 5.64%, 5.08%, 4.65%, 4.45%, 3.95%and 3.93% of the total variation, respectively (Table 2).

Table 2. Principal component analysis, eigenvalues and percentage variations of twenty five phenotypic traits of 131 cassava genotypes

Principal Component	**Characteristics**	Initial Eigenvalues		
		Total	% of variance	Cumulative %
1	Shape of central leaflet	3.27	13.07	13.07
2	Lobe margins	2.39	9.55	22.62
3	Colour of stem epidermis	2.04	8.15	30.76
4	Colour of stem exterior	1.74	6.97	37.73
5	Leaf colour	1.54	6.15	43.88
6	Orientation of petiole	1.41	5.64	49.53
7	Extent of root puduncle	1.27	5.08	54.61
8	Colour of root cortex	1.16	4.65	59.26
9	Root shape	1.11	4.45	63.70
10	Root taste	0.99	3.95	67.65
11	Cortex thickness	0.98	3.93	71.58
12	Distance between leaf scars	0.88	3.50	75.09
13	Colour of root pulp (parenchyma)	0.82	3.27	78.36
14	Texture of root epidermis	0.77	3.08	81.43
15	Average petiole length	0.66	2.64	84.07
16	Color of apical leaves	0.62	2.49	86.57
17	Cortex ease of peeling	0.57	2.26	88.82
18	Colour of end branches of adult plant	0.51	2.02	90.84
19	Flowering	0.46	1.83	92.67
20	Colour of stem cortex	0.42	1.66	94.35
21	Number of leaf lobes	0.35	1.40	95.74
22	Colour of leaf vein	0.34	1.35	97.09
23	Petiole colour	0.30	1.20	98.29
24	External colour of storage root	0.24	0.98	99.27
25	Average ratio of lobe length to lobe width	0.18	0.73	100.00

Phenotypic characterization

Cassava landraces analyzed revealed larger degree of morphological variations based on 25phenotypic qualitative and quantitative descriptors used (Table 1).The dendrogram obtained (Fig 3)using phenotypic characters separated the 131cassava genotypes into four major clusters (1, 2, 3 and 4) at similarity index of 0.5. Results presented in Table3 showed that 95cassava genotypes accounting for 72.5% of the accessions were grouped in cluster #1 (Table 3). Cluster #2had 21phenotypes representing 16% of the total number of entries. Cluster #3 and 4 had 4 genotypes (3.1%) and 11 genotypes (8.4%) respectively (Table 3).

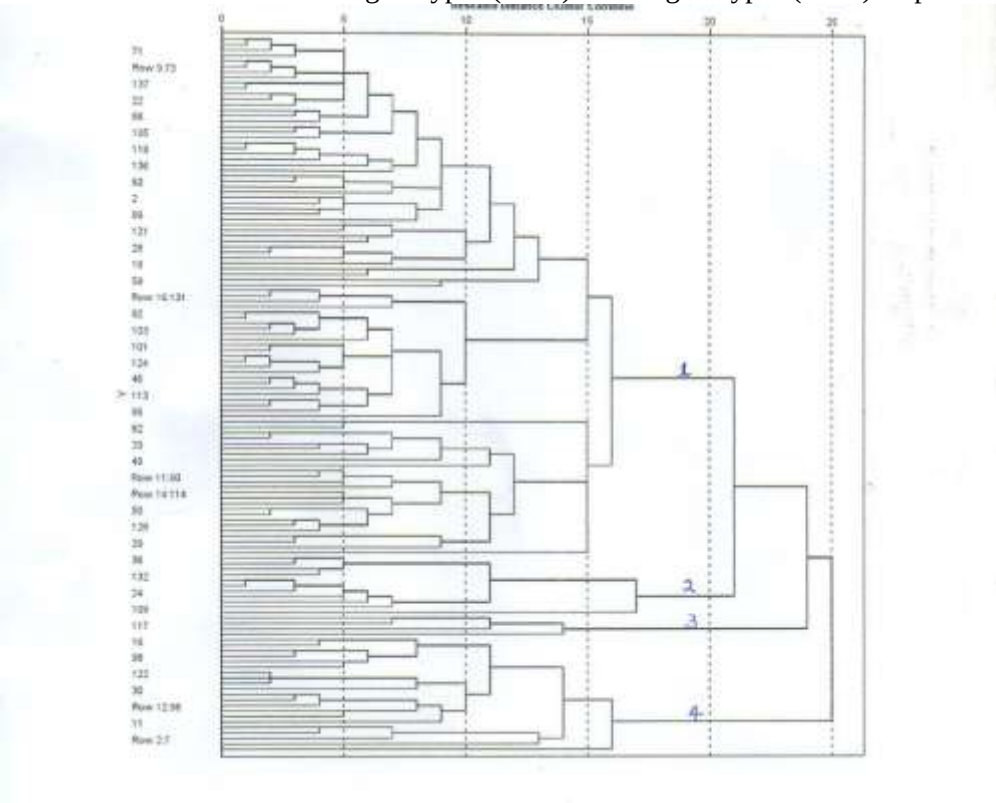

Figure 3: Dendrogramillustrating131Cassava genotypes based on average linkage (between groups) cluster analysis

Table 3: Names of cassava varieties, locations and the counties they were collected

CLUSTER #1			
Accession No.	**Variety**	**Location collected**	**County**
Row 14:114	Nyakanyamkago	Sigiria	Migori
Row 16:131	MM96/0039	Chakol	Busia
Row 13:104	Nyar-ICIPE	Sigiria	Migori
Row 10:83	Mygera	Rongo	Migori
Row 11:90	Adhiambo lera-002	Mtwapa	Kilifi
Row 4:23	Nyatanga	Rabuor	Kisumu
Row 5:33	Nyarkokaro	Rakwaro	Migori
Row 7:53	Amakuria	Masaba- Kehancha	Migori
Row 9:73	Busia-004	Busia	Busia
106	Agriculture-019	Maram	Homa bay
107	Nyatonge-002	Sigiria	Migori
108	Mygera-002	Sigiria	Migori

110	Obayo dak-003	Ranen	Migori
111	Otia	Sigiria	Migori
112	Agriculture-020	Ranen	Migori
113	Agriculture-021	Maram	Homa bay
115	Konono	Ranen	Migori
116	Agriculture-022	Pembe	Migori
118	Agriculture-23	Kitere	Migori
119	Agriculture-024	Sigiria	Migori
120	Unknown	Maram	Homa bay
121	Obaro dak-004	Nyamarere	Migori
124	Mufutu	Migori	Migori
126	Nyaodendo	Sigiria	Migori
127	NyarMaseno	Busia	Busia
128	MM96/4878	Busia	Busia
129	Yellow - 002	Chakol	Busia
130	MH95/0183	Chakol	Busia
134	Magana	Chakol	Busia
135	MH96/0031	Chakol	Busia
136	Migyera - 003	Chakol	Busia
137	Bwana Terana	Chakol	Busia
138	MH95/2480	Kolwa	Kisumu
100	Agriculture-018	Ranen	Migori
101	Bwong	Ranen	Migori
102	Obaro dak-001	Maram	Homa bay
103	Nyarkanyamkago	Ranen	Migori
105	Obar dak-002	Nyamarere	Migori
12	Agriculture-003	Rapogi	Migori
18	Katune	Kiboko	Makueni
19	KME-4	Kiboko	Makueni
20	Kazanzwara	Kiboko	Makueni
21	KBK-20	Kiboko	Makueni
22	Agriculture-004	Busia	Busia
25	Nyaeta	Kehancha	Migori
26	Wild cassava-001	Ranen	Migori
28	Kienyeji	Maram	Homa bay
29	Wild cassava -002	Rusinga island	Homa bay
34	Unknown variety	Kegonga-Kehancha	Migori
37	Buria	Maeta, Kehancha	Migori
38	NyaogutuNgalo	Maram	Homa bay
40	Nyagire	Maram	Homa bay
41	Agriculture 006	Opago	Migori
44	Unknown variety-003	Maram	Homa bay
45	Nyasega	Rakwaro	Migori
46	Nyasuna	Kokuro	Migori
47	Nyatanga-003	Opapo	Migori
48	Nyakakelo	Kanga	Migori
49	Obiero Abele	Ranen	Migori
50	Opoto	Uriri	Migori
51	Nyakasamwel	Awendo	Migori
52	Nyarkagutu	Ngothe	Migori
54	Agriculture-007	Suba; Kuria	Migori

55	Agriculture-008	Busia	Busia
56	Agriculture-009	Masaba; Kehancha	Migori
57	MM96/4466	Subukia	Nakuru
59	Busia-001	Busia	Busia
60	Busia-002	Busia	Busia
61	Machoberi	Kegonga;Kehancha	Migori
66	Agriculture-011	Kegonga; Kehancha	Migori
67	Agriculture -012	Awendo	Migori
71	Agriculture -013	Dede	Migori
74	Nyarkogutu-002	Ngothe	Migori
75	Nyarkadera	Kadera	Migori
76	Agriculrure-014	Uriri	Migori
78	Unknown variety-004	Kehancha	Migori
80	Agriculture-015	Busia	Busia
81	Agriculture-016	Rongo	Migori
82	Waite-002	Kegonga; Kehancha	Migori
85	Achuth	Uriri	Migori
86	Nyanchama	Rongo	Migori
87	Nyatanga-004	Rongo	Migori
88	F-19	Mtwapa	Kilifi
91	Katsuhanzala	Mtwapa	Kilifi
92	Kasukari	Mtwapa	Kilifi
93	Karembo	Mtwapa	Kilifi
94	Mtwapa-002	Mtwapa	Kilifi
95	Tajirika	Mtwapa	Kilifi
97	Kibanda meno-003	Mtwapa	Kilifi
99	MM96/0067	Mtwapa	Kilifi
10	Agriculture-001	Ranen	Migori
2	Mzungu	Mida Creek	Kilifi
3	Miida	Mida Creek	Kilifi
6	Mbale-002	Mida Creek	Kilifi
9	Mary Kaluorore	Ranen	Migori
CLUSTER #2			
Row 15:123	Rateng	Pembe	Migori
Row 12:96	Agriculture-017	Mtwapa	Kilifi
Row 6:43	Odiero	Rongo	Migori
Row 2:7	Agriculture-001	Ranen	Migori
122	Ratena	Nyamarere	Migori
13	Yellow-001	Rapogi	Migori
14	Kamgundho	Rapogi	Migori
16	KBK-4	Kiboko	Makueni
17	KBK-21	Kiboko	Makueni
30	Nyarkawuor	Uriri	Migori
31	Wild cassava-003	Kendu bay	Homa bay
32	Agriculture-005	Maeta-Kehancha	Migori
42	Unknown variety-002	Rakwaro	Migori
62	AdhiamboLera	Awendo	Migori
72	Busia-003	Busia	Busia
77	Nyakasani	Awendo	Migori
89	Mtwapa-009	Mtwapa	Kilifi
98	MM96/0067	Mtwapa	Kilifi

11	Selele rachar	Rapogi	Migori
4	Mbale-001	Miida Creek	Kilifi
8	Nyaranen	Ranen	Migori
CLUSTER #3			
Row 3:15	Kasukali	Kiboko	Makueni
117	Toji	Kendu bay	Homa bay
58	Agriculture-010	Kegonga, Kehancha	Migori
68	Madam	Opapo	Migori
CLUSTER #4			
Row 8:63	Unknown variety-003	Rakwaro	Migori
109	Selele-007	Sigiria	Migori
125	Selele-009	Ranen	Migori
132	Fumbachai	Chakol	Busia
24	Selele-002	Rongo	Migori
27	Selele-003	Maram	Homa bay
35	Selele-004	Rakwaro	Migori
36	Nyatanga-002	Uriri	Migori
39	Selele-005	Maram	Migori
64	Selele-006	Busia	Busia
84	Nyasuna	Masaba, Kehancha	Migori

Discussion

Numerical taxonomic studies are important for discovering and documenting new character and character states (Rahman, 2013). Cluster analysis (CA) and principal component analysis (PCA) are two techniques commonly used in numerical classification (Sonibare *et al*., 2004). PCA is usually used as an exploratory tool in systematic. There are as many components as original variables, and these components are linear combinations of the original variables. Most of the variance is usually summarized by the first few components, and PCA thus reduces a larger number of variables to fewer variables, which are often easier to interpret and is thus described as a dimension reducing method (Rahman, 2013). Cluster analysis (CA) is an exploratory tool for classifying objects with no statistical assumptions about the data. Cluster analysis produces a hierarchical classification of entities (taxa) based on the similarity matrix. Results are usually presented in the form of trees or dendrograms (Henderson, 2006).

In this study eigenvalues and percentage variations of the principal component analysis were evaluated. Eleven principal components accounting for 71.58% of the total variation among the genotypes were identified. The PCs were: Shape of central leaflet, lobe margins, colour of stem epidermis, colour of stem exterior, leaf colour, orientation of petiole, extent of root puduncle, colour of root cortex, root shape, root taste and cortex thickness. Similar studies have been carried out in other regions. Studies carried out in Sierra Leone (Karim *et al* 2020) identified a total of seven principal components (PCs) in the qualitative and four PCs in the quantitative trait sets accounted for 79.03%and 72.30% of the total genetic variation in 102 cassava genotypes, respectively. In the same study five cluster groups were identified based on the qualitative agronomic traits. The estimation of descriptive statistics of 25 different morphological traits studied in the present study revealed the existence of morphological diversity among cassava landraces. From the small sample of 131 genotypes it was possible to characterize them into four clusters with a majority (72.5%) falling into one cluster. This signifies that a large population of cassava grown in Kenya have similar characteristics as only 27.5% are characterized in the other three clusters. This makes it difficult for farmers to select the distinctive planting materials. These results are similar with those obtained in Brazil (Tiago *et al*, 2020). Among the 45 cassava ethno-varieties studied, they presented 97.35% polymorphism, which showed that there were morphological divergence between the evaluated samples (Tiago *et al*., 2020).

The three wild cassava accessions included in this study were characterized in clusters where cultivated cassava were identified. Wild cassava – 003 (entry 31) was classified in cluster #2 while Wild cassava – 002 (entry 29) and Wild cassava – 001 (entry26) were classified in cluster #1. These results differed with the findings of Dissanayake *et al*., (2019) in Sri Lanka who carried out morphological assessment of cassava cultivars and established that the leaf morphology of wild-accessions and landrace cultivars were significantly different from the rest of the cultivars. Stem morphology among the cultivars was significantly different mainly by the mean inter-nodal length of the stems whereas wild-accession cultivars were significantly different from the rest by the diameter of the stems. In this study, it was expected that the wild cassava genotypes would be clustered in a distinct group. In Indonesia, details of 14 morphological characteristics for 29 cassava genotypes were used in cassava landrace characterization. It was established that almost all genotypes had purple petiole color and horizontal orientation, smooth lobe margin, and seven lobes (Ridwan *et al*., 2022). In Burundi, Niyonzima *et al*., (2021) assessed landrace cassava morphological traits and noted that stem, root and leaf traits distribution differed among cassava landraces.

The TMS cassava varieties released by KALRO and named *Agriculture* by farmers were coded Agriculture – 001 – Agriculture 021. The genotypes appeared in clusters 1, 2 and 3; signifying that the varieties that were in different clusters were different varieties. This is a first step in the identification of these genotypes. There were also varieties whose names the farmers did not know (*Unknown variety*). This is a common occurrence especially for farmers who are planting cassava for the first time or who are recently introduced to cassava farming. For them the name of the variety is not important. This observation also applied to the genotypes collected from Mtwapa (Kilifi County). It was expected that this study would shed light on their identities. Further genetic studies need to be carried out to correlate the phenotypic and genotypic traits for proper characterization.

Conclusion

This study highlighted the phenotypic variability within the cassava genotypes collected. Despite the variability found within the germplasm, it is concluded that cassava phenotype base in Kenya is narrow as it was revealed that 72.5% phenotypes were clustered in one group. Genotypes with very close morphological characteristics such as Adhiambo lera in Migori (cluster #1) and Adhiambo lera (cluster #2) in Mtwapa should be considered as putative duplicates, hence, need to be pooled together as one cultivar.

Recommendations

Future studies on phenotypic characterization should focus on the 11 phenotypic traits that accounted for 71% cumulative variation. The application of phenotypic descriptors in identification of cassava landrace germplasm should be backed by the use of molecular markers (genetic characterization), since the former alone does not reveal much diversity due to the effects of the environment on quantitative traits.

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Conflict of Interest

"The author(s) declare(s) that there is no conflict of interest." There was no role of the funding sponsors 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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Local Farmers' Perception in Managing Fall Armyworm, *Spodoptera frugiperda,* in Maize Crop in Northwest of Benin

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Abstract
The fall armyworm, *Spodoptera frugiperda* (Lepidoptera: Noctuidae) is a major insect pest which has a potential to inflict widespread damage across multiple crops since its arrival in Africa. This study assessed local farmers' perception and associated alternative methods against the infestation of fall armyworm in maize fields in Northwest Benin. Data were collected through surveys with 180 randomly selected farmers. Results indicated that all interviewed farmers have knowledge of natural enemies especially predators to control the infestation of the fall armyworm. We noted that 99% of interviewed farmers associated the high insect pest occurrence with the extreme weather and climatic conditions and only 48.3% linked the high infestation degree with the density of maize yield. The use of chemical insecticides is the most applied method by all farmers to control the pest, though some of them (14.4%) valued alternative management practices based on their indigenous knowledge in the control of the fall armyworm. Additionally, the results showed positive learning outcomes from the workshops on alternative pest control management attended by the local farmers. Those workshops act as a catalyst in improving the knowledge on alternatives methods. This study provides a baseline in capitalizing local management practices deemed effective by farmers in reducing fall armyworm infestation.
Keywords: Alternative methods, insecticides, climatic conditions, local knowledge

Introduction

Maize, *Zea mays* L., (Poaceae) is an important staple food and cash crop in Sub-Saharan Africa (SSA). It has a significant nutritional value and economic importance in the diets of the Beninese population (FAO, 2017; MAEP, 2017; Bangale, 2019). Yet, the production of this cereal faces several challenges, including abiotic and biotic constraints. Among the biotic factors, the recent emergence and spread of the fall armyworm, *Spodoptera frugiperda* caused devastating damage to the maize crop (Wan et al., 2021). The fall armyworm, *S. frugiperda* is a major insect pest species (Senay et al., 2022) originating from the American continent (Early et al., 2018), where it causes large damage to multiple crops (Knipling, 1980; Montezano et al., 2018). In Africa, the fall armyworm was first reported in Southwestern Nigeria in 2016 and shortly thereafter in northern Benin and Togo (Goergen et al., 2016). Since then, the fall armyworm has started to spread rapidly, becoming the most damaging pest for maize production (Goergen et al., 2016; Rwomushana et al., 2018). The fall armyworm causes harm in the seedling stage of maize plants, which are most appreciated by the pest (Akinyemi, 2021). Due to its rapid expansion, the fall armyworm poses an imminent threat to the food and nutrition security, as well as the livelihoods, of hundreds of millions of

farmers and their families in Sub-Saharan Africa (Prasanna et al., 2018). According to an evidence note published in September 2017 by the Centre for Agriculture and Bioscience International (CABI), in the dearth of adequate control methods, the fall armyworm has the capacity to cause maize yield losses ranging from 8.3 to 20.6 million metric tons per year in only 12 of Africa's maize producing countries (Prasanna et al., 2018). Almost all major damage by fall armyworm has been recorded for maize production, while little to no damage has been recorded for other crops in Africa (Rwomushana et al., 2018).

In Benin, the fall armyworm has been reported by the government to mainly cause injuries to maize crops (MAEP, 2016). According to surveys conducted in 2016, over 395,000 acres of maize suffered damage, which led to an estimated loss of 415,000 tons (30% of the country's production) (MAEP, 2016). This loss in productivity presents a threat to the country's food security and economy (Houngbo et al., 2020). Therefore, it is urgent to develop alternative control options that can be incorporated into a sustainable integrated pest management approach to control the spread of the fall armyworm (Harrison et al., 2022).

In recent years, research has pointed out the need to supplement the local knowledge of farmers with sustainable and environmentally friendly alternatives in pest management (Wyckhuys and O'Neil, 2007). Farmers have local knowledge and practices that provide opportunities for alternative and sustainable pest management (Kansiime et al., 2019). However, their knowledge and management strategies for the fall armyworm have been poorly documented since its first appearance in Africa (Houngbo et al., 2020). Limited efforts have been made by researchers in Ethiopia (Kumela et al., 2019), Ghana (Ninson et al., 2020), Burkina Faso (Ahissou et al., 2022), and Benin (Houngbo et al., 2020) to specifically address farmer's knowledge in their studies. Yet, these studies failed to provide specific information regarding the local farmers' understanding of the invasion of fall armyworm, their knowledge and their capacity to develop integrated pest management. This study is an attempt to understand local perception on the invasion of the fall armyworm, the farmers' knowledge on the fall armyworm, different methods used to handle the infestation, and the farmer's ability to develop and implement integrated pest management methods. In this study we refer to farmers' understanding as how they perceive the invasion of fall armyworm and the related damage caused by the pest. The local knowledge of farmers is described here as a body of knowledge they have been using locally and transmitting orally through generations. Farmers' knowledge is also understood as how much they know, control, and manage the pest. As argued by Van Mele et al. (2001), one of the main obstacles to the implementation of pest control programs is the lack of information on farmers' knowledge, understanding and management practices. This research explores the existing management practices applied by farmers against the fall armyworm and test their effectiveness. The objective is to build the understanding around farmers' knowledge and alternative methods associated against the spread of the fall armyworm in Northwest Benin.

Material and Methods

Study Area

This study was conducted in the department of Atacora (10° 45' 0" N and 1° 40' 0" E) and particularly in the municipalities of Pehunco (10° 13' 45" N and 1° 59' 58" E), Natitingou (10° 18' 14" N and 1° 22' 46" E) and Tanguiéta (10° 37' 21.919" N and 1° 15' 54.126" E) (Fig. 1). These are some of the most important maize-growing areas, with maize and sorghum cropping systems linked with animal production. The study area is characterized by a Sudanian-type climate with a rainfall variation between 900 and 1100 mm/year (spread over an average of 71 days) and an average monthly temperature ranging from 24°C to 31°C (PNA, 2022). The vegetation is composed of wooded savannahs and forest galleries (PNA, 2022). This study area is chosen mainly because of its climatic condition, which is favorable for the spread and high prevalence of *S. frugiperda* (Senay et al., 2022; Goergen et al., 2016).

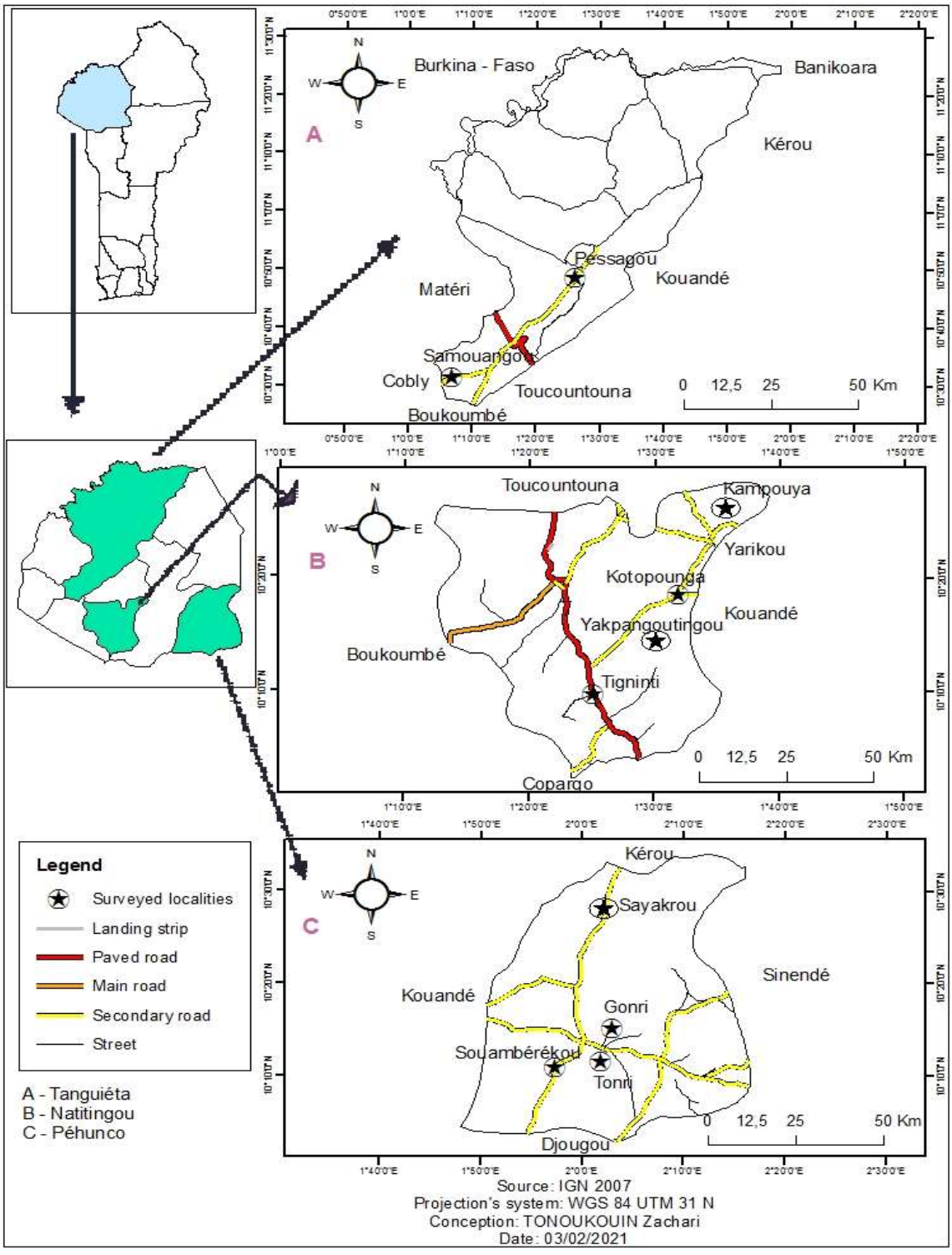

Figure2: Map showing the study area

Data Collection

The survey was conducted from July to September 2020 in the selected municipalities. The survey municipalities were chosen on purpose based on reports of the fall armyworm's appearance via official organization websites and news media (Kumela et al., 2019). In each municipality, 60 local maize growers were randomly selected for interviews from sampling lists obtained from the AGRO-ECO Project. A total of 180 maize growers were subjected to interviews in Pehunco, Natitingou and Tanguita. A semi-structured questionnaire was administered to all selected farmers. The questionnaires were developed in French and discussed in both French and local language (Batonum; Waama; etc.) during face-to-face interviews with individual farmers. A local translator in each municipality was recruited to assist in translating the questionnaires to farmers that could not understand French. The objective of the study was explained to all selected farmers and verbal consent was given prior to their participation to the interview. The information collected included farmers' demographic and socio-economic profiles, farmers' perceptions of damage caused by the fall armyworm, farmer's knowledge of the pest, and the management practices used for its control. The technique of free lists (Brewer, 1995) was used to assess local knowledge used by farmers for pest management practices. The degree of severity of fall armyworm infestation as perceived by farmers was assigned as a value using five-point rating scale: (1) no infestation; (2) low infestation; (3) moderate infestation; (4) high infestation and (5) very high infestation. Farmers were asked to describe the fall armyworm infestation levels and their perceptions of factors that were associated with the severity of infestations. Methods for controlling cereal insect pests in general, and particularly the fall armyworm, with associated knowledge as well as alternative techniques were also recorded. Afterwards, farmers were asked to describe the criteria used to decide when and how to manage the pest. Criteria are understood here as either weather, unusual observed change in the crop, the extent of damage caused by the pest and the season.

Data Analysis

Survey data were summarized in an excel spreadsheet and descriptive statistics (means and percentages) were computed using the Statistical Package for Social Sciences 21.0 (SPSS). Percentages of farmers with a similar response to a question were calculated based on the total number of farmers who responded to each question. Moreover, the data were analyzed following two statistical analysis tools, such as Chi-square and one-way analysis of variance (ANOVA). These analyses were conducted to assess the correlation and differences between variables such as farmers' demographic and socio-economic profiles, farmers' perceptions of damage caused by the fall armyworm, farmer's knowledge of the pest, and the management practices used for its control. The level of significance was set at 0.05. Additionally, Cramer's V test was run based on Chi-square to measure the strength of the correlation between two variables in percentage and their maximum possible variation.

Results

Socio-demographic characteristics of the farmers surveyed

The socio-economic profile of farmers surveyed in the three municipalities is summarized in Table 1. The majority (88%) of the farmers surveyed were men. About 72% of the farmers surveyed have more than 15 years of experience in maize production. Of all the respondents, 59% produce maize covering an area of 0.5 to 2 ha, and 25% produce maize covering an area between 2 and 5 ha. Besides maize, a diversity of crops were grown at differing production scales in the different communes. 82% and 78% of the respondents, respectively from the communes of Pehunco and Natitingou, adopted maize as their main crop.

Table 1: Profile of maize farmers in the three communes surveyed

Variables		**Pehunco (N = 60)**	**Natitingou (N = 60)**	**Tanguiéta (N = 60)**	**Total (N = 180)**
Age (average)		33.4±8.5a*	42.3±14.7b	37.7±15.9ab	37.8±13.8
Household size		7.6 ± 4a	7.6 ± 4.2a	5.8 ± 3.7a	6.7 ± 4
Agricultural assets		6.6 ± 4.1a	5.6 ± 3.2a	4.6 ± 3.2a	5.1 ± 3.2
Parameters			Percentage %		
Gender	Male	93	75	95	88
	Female	7	25	5	12
Religion	Christian	28	57	13	33
	Muslim	48	10	3	20
	Animist	24	33	84	47
Education		48	50	35	44
Ethnic group	Waama	2	65	43	37
	Batonum	80	2	0	27
	Kountimba	0	0	53	18
Farmers in terms of years of experience	1 – 5 years	12	5	12	9
	6 – 10 years	10	10	12	11
	11 – 15 years	12	2	10	8
	More than 15 years	67	83	67	72
Surface area of maize (2019/2020)	Less than 0.5 ha	2	18	0	7
	0.5 – 2 ha	42	70	65	59
	2 – 5 ha	33	12	30	25
	5 – 7 ha	18	0	3	7
	More than 7 ha	5	0	2	2
Main crop	Maize	82	78	55	72
	Sorghum	0	7	0	2
	Millet	0	5	0	1.5
	Rice	1.5	0	3	1.5
	Soybean	15	0	2	6
	Yam	0	5	0	2
	Cotton	1.5	3	38	14
	Vandzou	0	2	0	0.5
	Sesame	0	0	2	0.5

***** Average ± standard deviation; values of the same line followed by identical letters are not significantly different at 5% (one-way ANOVA followed by Tukey's test*).*

Knowledge of Maize Insect Pests and Estimation of the level of Severity of *S. frugiperda* attacks

The evaluation of farmers' perception on the severity level of fall armyworm attacks showed significant differences ($p < 0.05$) between the three communes (Table 2). About 52.8% of farmers mentioned high levels of fall armyworm infestations, ranging from 51 to 80%.

Table 2: Severity estimation of *S. frugiperda* attacks by farmers

Attack severity scale	**% of farmers**				**Cramer's V**
	Pehunco (N = 60)	Natitingou (N = 60)	Tanguieta (N = 60)	Total (N=180)	
No infestation (0%)	0	0	0	0	0.276*
Low (1-20%)	0	0	0	0	
Moderate (21 – 50%)	20	5	0	8.3	
High (51 – 80%)	61.7	50	46.7	52.8	
Very high (81 – 100%)	18.3	45	53.3	38.9	

*Significant at $p < 0.05$
X^2 (Chi 2) = 27.325 (ddl=4; N=180); P-value = 0.000; α= 5%.

Several causes were identified by farmers to explain the levels of fall armyworm infestation (Fig. 2). Among others, the extreme climatic events (drought, storm, etc.) and the use of chemical pesticides have been identified as the first factors explaining the high infestation of fall armyworm in the study area.

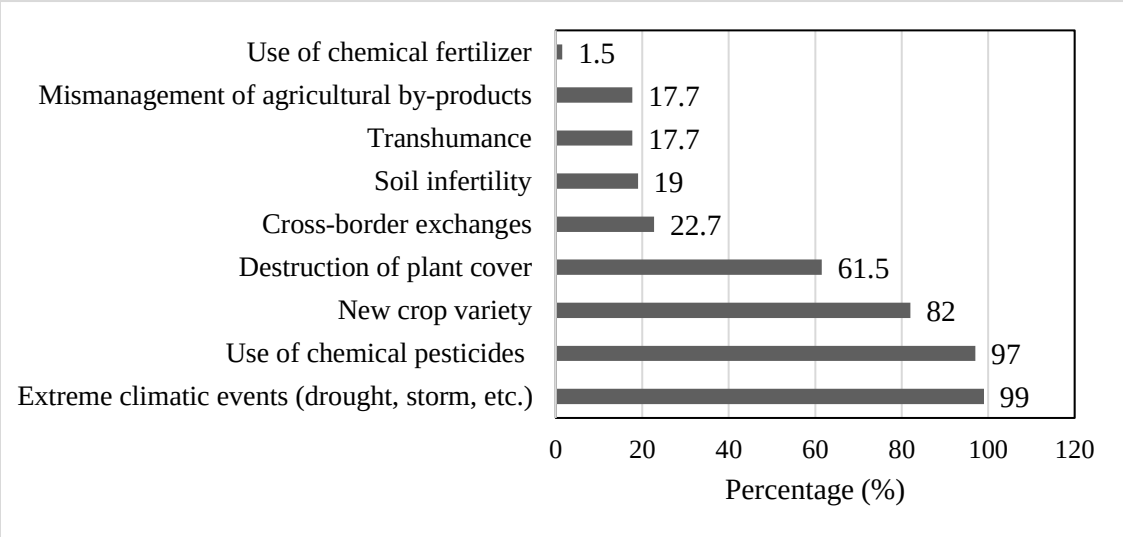

Figure 3: Perception of farmers on factors allowing the proliferation of fall armyworm

Knowledge of fall armyworm enemies/predators by farmers

Farmers mentioned several natural enemies and predators in their maize fields (Table 3). Almost all interviewed farmers have repeatedly reported the presence of predators in their fields and have expressed knowledge on how to use those predators to control the fall armyworm infestation. By recognizing the roles played by predators such as birds, frogs, lizards, and chicken in regulating the population of fall armyworm, farmers intentionally introduce those predators as their local solution to stop the spread of the pest.

Table 3: Enemies/predators of fall armyworm, mentioned by farmers

Predator names	**Local name (Waama; Batonum)**	**% of farmers**		
		P	NT	T
Invertebrates				
Dermaptera: earwig	Sonahun; Dodoniyan	100	100	100
Hymenoptera: ants	Toukounan; tankinga	100	100	100
Mantoptera: praying mantis	Koxawounga; Gouhoudouma	100	100	100
Coleoptera: beetle	Krokroré; Mangbi	100	100	100
Hemiptera: bedbug (*Podisus* sp.)	Pekne; Abrakounkoungou	75	90	100
Arachnid: scorpion	Nangou; Niyan	100	100	100
The vertebrates				
Birds	Nonnonfa; Kinkinyami	100	100	100
Frogs	Tcharbou; Frerou	100	100	100
Lizards	Wouroukou; Gninnou	100	100	100
Chickens	Koxa; Gouwa	100	100	100

P=Pehunco; NT = Natitingou; T = Tanguieta

Influence of Cropping System on fall Armyworm Infestation

Farmers reported the high density of maize crop as the main cause of high infestation by fall armyworm (Fig. 3). The pest infestation is highly observed in mono-cropping systems as mentioned by farmers. For instance, growing one sweet variety of corn in the same field year after year is identified as a main driver of the spread of the fall armyworm. Additionally, farmers reported other related factors that increase the degree of fall armyworm infestation as shown below.

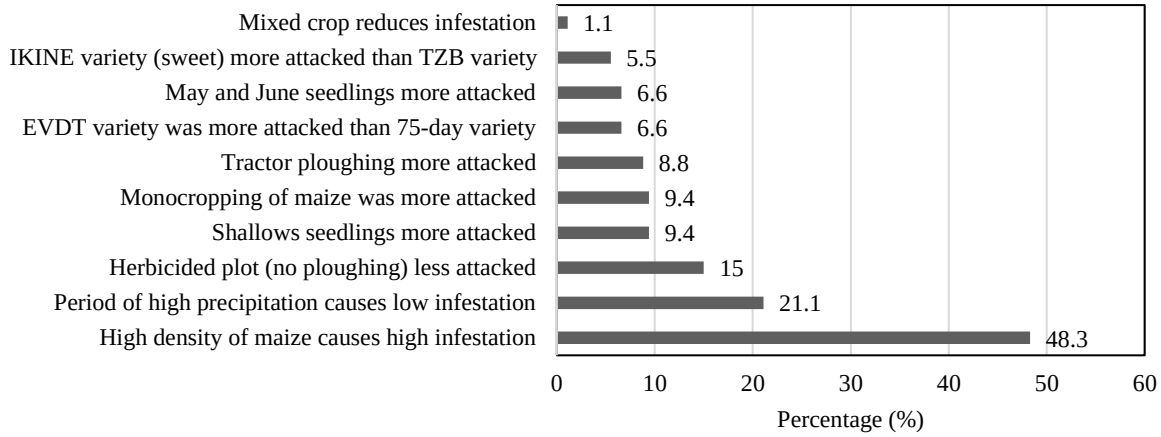

Figure 4: Percentage of the factors influencing the spread of fall armyworm.

Management practices of fall Armyworm Infestation by farmers

Some 60% of all interviewed farmers have reported no control measure against the spread of fall armyworm infestation (Table 4). Farmers in Pehunco (95%) have not developed any pest control management practices, while in Natitingou (45%) and Tanguieta (54%) farmers have respectively used synthetic chemical insecticides and alternative methods as first management practice of controlling the pest.

Table 4: Management practices of fall armyworm infestation, mentioned by farmers

Management Practices	P (N=60)	Nt (N=60)	T (N=60)	Total (N=180)
No control measures (%)	95	38.3	46.7	60
Use of synthetic chemicals (%)	0	45	31.7	25.6
Super Lambda (Lambdacyhalothrin)	0	43.3	0	14.4
Thalis 112 EC (Emamectin benzoate, Acetamiprid)	0	0	31.7	10.6
Emacot 050 WG	0	1.7	0	0.6
Use of biopesticide (%)	0	0	0	0
Thalis + neem leaf extract	1.7	0	0	0.6
Soapy water (Omo)	3.3	16.7	53.3	24.4
Neem extract + pepper + soap	0	0	53.3	17.8
Neem + papaya + garlic extracts	0	0	31.7	10.6
Other extracts used against other insect pests (%)				
Pema extract[a] (local name in Waama)	0	5	0	1.7
Extract of *Euphorbia* sp.	0		0	1.7
Extract from Tuandi[a] (local name in Kountimba)	0	0	1.7	0.6

[a] cited and used by farmers against other insect pests but unknown to the authors

P=Pehunco; NT = Natitingou; T = Tanguieta

There is a significant correlation (p=0.000; Chi2 = 300.817) between the choice of specific pest management practice by farmers and their communes of residence (Table 6). The location of farmers and their farms determine the type of management practice implemented. For example, farmers in Pehunco (58.3%) reported that they are not aware of any effective pest control product, while farmers in Natitingou (22%) claimed about the effectiveness of *Lambda Super* in controlling fall armyworm infestation.

Participation of Farmers in pest Management Trainings

The high participation (97%) of farmers from Tanguiéta in pest management workshop sessions has direct implications on their choice in pest management products. Almost all of them have applied botanical pesticides as a local way of controlling the pest. This is in contrast to farmers from Natitingou and Pehunco, who never used any botanical pesticides (Table 5). Accordingly, most farmers in Natitingou and Pehunco have never participated in workshop sessions on pest management practices.

Table 5: Participation of farmers in insect pest management trainings

Participation in training and control technologies	**Percentage (%)**		
	P	**Nt**	**T**
Participation and topics covered during the training			
Participation in training	4	4	58
Recognition of insect pests	3	1	58
Use of chemical pesticides	3	1	32
Preparation and use of botanical pesticides	3	4	58
Recognition of natural enemies	0	1	32
Control technology taught against insect pests			
Use of chemical insecticide	4	2	58
Use of ashes	0	1	32
Use of botanical insecticide (neem; papaya; garlic)	3	3	58
Use of soapy water	0	1	32
Fall armyworm control technology or innovation adopted			
No control measures	57	23	28
Use of chemical insecticide	0	27	19
Blend of chemical and botanical insecticide	1	0	0
Use of soapy water	2	10	32
Neem leaf extract + garlic + ash	0	0	13
Neem leaf extract + pepper + soap	0	11	19
Mixture of Thalis 112 EC + neem leaf extract	1	0	0
Sowing date offset	0	12	0
Staggered sowing	0	10	0
Intercropping	20	8	10

P = Pehunco; Nt = Natitingou; T = Tanguiéta

Discussion

In this study, all the surveyed farmers mentioned the fall armyworm as being the most important current pest of the maize crop. Similar results were obtained in Ghana (98.1%) and Zambia (98.6%) (Rwomushana et al. 2018). In support, the study of Kouanda (2020) showed that 96% of farmers surveyed in Burkina Faso cited maize as the most attacked by fall armyworm caterpillars. This is due to the high density of maize crop which is identified in this study as one of the main factors attracting the fall armyworm. The results of this study shows that a high seedling density is likely to cause high infestation by fall armyworm. Farmers argued that seedlings in lowlands and early sowings are frequently attacked by fall armyworm. In opposite, evidence from Harrison et al. (2019) in North America has indicated that early sowings are among

successful strategies to control the spread of fall armyworm. The author stated that the results would likely be very different in African context due to the variability in climate conditions (Harrison et al., 2019). Differences in the degree of infestation depending on the variety of maize grown were also mentioned by farmers.

Other crops such as sorghum, millet, rice, soybeans, and cotton are also invaded by *S. frugiperda* caterpillars. These crop species would contribute to the spread and the persistence of the pest in Northern Benin. These crops associated with maize have shown a significant impact in reducing the spread of the fall armyworm. Farmers in Pehunco have adopted intercropping systems (maize and beans/groundnuts) and is proved as being efficient in the control of fall armyworm. The study conducted by Letourneau et al. (2011) revealed that intercropping systems adopted by farmers have significantly reduced pest insect populations, increased natural enemy populations, and reduced crop damage. The results also show the significance of using insecticides in reducing the spread of the pest. Chemical insecticides are largely used by farmers to control the spread of fall armyworm even though some of them claim to apply alternative methods because of the high cost of chemical insecticides and the ineffectiveness of chemicals control. Similar results were obtained in Burkina Faso (Kouanda, 2020), Ethiopia and Kenya (Kumela et al., 2019), where many maize growers mainly used chemical insecticides to control the infestation by fall armyworm. Many other studies have discussed the efficiency (Aniwanou et al., 2021) or not (Adeye et al., 2018) in using chemical insecticides. According to Boukari et al. (2022), the application of insecticides by farmers does not guarantee long-lasting protection against fall armyworm infestation. Yet, the application of insecticides comes with some challenges as revealed by our study. The pests that caused damage are more active early in the morning and late in the afternoon or evening. Therefore, insecticide applications by farmers that do not target these times (early morning or late afternoon) of the day render treatments or other control arrangements ineffective. Also, one of the difficulties of using insecticides to control the fall armyworm is that the pests tend to hide inside the whorls and reproductive organs of the host plant, where it is difficult for insecticides to reach.

The degree of fall armyworm infestation in both farming systems is associated to factors including the use of chemical pesticides, droughts, the introduction of new crop varieties, destruction of plant cover, extreme climate events, etc. Research by Houngbo et al. (2020) reported that *S. frugiperda* attacks were more severe on new varieties of crops than other ones. In all three communes, 92% of interviewed farmers (92%) reported that heavy rains reduce the degree of attacks by *S. frugiperda* caterpillars on maize crops. Heavy rains can interrupt the life cycle of the fall armyworm by washing away the eggs from the leaves. They can also drown young larvae in maize whorls, causing high mortality rates. In the region of Honduras in America, farmers linked the degree of fall armyworm infestation to the variations in weather and climatic conditions (Wyckhuys and O'Neil, 2007). Peer and Westbrook (1995) and Vanhuis (1981) also argued that heavy rains negatively impact the spread of the fall armyworm population.

Interviewed farmers have knowledge on natural enemies or predators of the fall armyworm. They recognized the role played by birds in reducing the spread of the pest on their farms. As reported by Wyckhuys and O'Neil (2007), birds are crucial in controlling the invasion of fall armyworm in maize yields in Honduras. Birds are known as fall armyworm predators (Luginbill, 1928) and their predation potential on Lepidoptera could reach high levels in some agroecosystems (Hooks et al., 2003). In our study, interviewed farmers believe more in the capacity of vertebrate species to effectively reduce the spread of fall armyworm such as chicken, bird, etc. This limits their ability to explore the predation ability of invertebrates such as ants, spiders, and earwigs. Consequently, limited local farmer's knowledge of these predators' ability in reducing the spread of fall armyworm could lead to a gap in conservation efforts addressing the pest management (Wyckhuys and O'Neil, 2007).

Farmers in the study used botanical pesticides as effective management practices of fall armyworm. Houngbo et al. (2020) reported the existence of significant relationships between farmers' management practices, their knowledge, and their involvement in research studies in Benin. Creating a safe space where farmers become familiar with alternative methods of handling the spread of fall armyworm is crucial in sustainable management of pests. The use of botanical insecticides is recommended as an alternative to chemical insecticides which can lead to environmental disruption (Arya and Tiwari, 2013). Farmers use alternative method of pest management which they claim to be part of their local solutions to control the pest. As such, soapy water, neem and papaya extracts, pepper, garlic and some combination of neem extracts and chemicals insecticides are applied by farmers in the fields. Evidence from Aniwanou et al. (2021) showed that the use of local soap called ''*Palmida*'' has provided effective control of fall armyworm infestation in Benin. Plants such as neem, tamarind and shea have also been mentioned by farmers as an alternative method against fall armyworm (Houngbo et al., 2022). Moreover, other botanical extracts such as Pèma extract (local name in Waama), *Euphorbia* sp. and Touandi (local name in Kountimba) have been used by farmers against other insect pests included the fall armyworm. Yet, those substances require further scientific analysis to test their long-term effectiveness against the pest (Hruska, 2019).

Participation of farmers in alternative methods of pest control workshops has significantly improved their knowledge on alternatives methods of reducing of fall armyworm infestation. That is the case of Tanguiéta municipality where a high record of the use of local knowledge and alternative methods were developed by farmers. As compared to farmers in Tanguieta, those in Natitingou and Pehunco have not participated in any pest management training. As argued by Wyckhuys and O'Neil (2007), there is a huge gap difference in knowledge and alternative methods developed against the fall armyworm by trained farmers and untrained ones. There is a need for further research to address more oriented training of farmers in alternative methods of fall armyworm control where needed.

Conclusion

This study addressed local knowledge of farmers and associated alternative methods against the spread of the fall armyworm in maize fields in Northwest Benin. Farmers have limited knowledge of natural enemies and use some few predators as a local solution of stopping the spread of the fall armyworm. Plus, the application of chemical insecticides is the most adopted method by farmers in controlling the fall armyworm infestation. According to some of them, the use of local practices based mainly on botanical and plant extracts, e.g. the application of soapy water, ash, peeper, and garlic, are more effective against the pest. Intercropping systems (maize and beans/groundnuts) adopted by farmers is proved as being efficient in the control of fall armyworm. The participation of farmers in alternatives methods of pest management workshops has a significant impact on their knowledge to reduce the fall armyworm infestation. Evidence from this study confirms that further research is needed to optimize and uniformly adapt the practices deemed effective by farmers to each local context in reducing of fall armyworm infestation.

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Microbiological Status and Physico Chemical Parameters of Fish and Water Samples across Selected Farms in South East Nigeria

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Abstract
A grand total of 12 water samples was collected from 4 different fish ponds (Ebenebe, Amansea, Amaenyi and Okpuno) all located in South East of Nigeria 3 samples each from concrete ponds between March and June 2021. The recorded mean physicochemical values for pH, temperature, electrical conductivity (EC), total dissolved solid (TDS), dissolved oxygen (DO), biological oxygen demand (BOD), turbidity and water hardness were within WHO guidelines for water quality. There was no significant difference ($p>0.05$) in the total bacterial load in fish samples from the stations. The mean heterotrophic bacterial counts and total coliform counts in fish samples ranged from $108.33\pm11.06x10^4$cfu/g to $105.00\pm36.86x\ 10^4$cfu/g, $97.66\ x\ 10^4$cfu/g and $79.00\pm16.52x\ 10^4$cfu/g, respectively. The bacterial isolates identified were; *Eschericia spp, pseudomonas spp, Vibrio spp and Salmonella spp,* Staph. Spp, Acinetobactercal coaceticu, Yersinia spp *and plesiomonas spp.* The study revealed that the pond waters involved was contaminated by bacteria, which might affect the growth of fish in the pond and may become harmful to human and also in food-chain over time It is therefore recommended that fishes be properly cooked and the microbiological analysis and physicochemical examination of wastewater or fish for signs of possible infections should be conducted on a regular basis.
Keywords: Fish ponds, Microbiological, Water, Physicochemical parameters, Fish, WHO

Introduction

Background to the Study

Fish is the most important single source of protein providing 16% of the animal protein consumed by the world's population (FAO, 2009). There is increased demand for fish in Nigeria especially the south east which cannot be met by the capture fisheries with stocks stagnating due to over exploitation. The alternative has been the growth in aquaculture with many people turning to farmed fish which is believed to be of better quality whereas it is documented that aquatic environments harbour many bacteria (Novotny, 2004). The use of organic waste for the fertilization of ponds is also a source of pathogenic organisms that may be transmitted to humans via products of aquaculture.

Water from underground sources such as a well is pumped and stored in storage tank which is used to refill the ponds when the water is to be changed. The feed used for fish in these ponds contain organic materials and introduces a wide variety of microorganisms into the ponds. Omojowo and Omojasola (2013) isolated six bacterial pathogens: *E. coli, E. coli 0157:H7, Shigella dysenteriae, Staphylococcus aureus, Salmonella typhi,* and *Aeromonas hydrophila* from Cow dung used as feed in the fish pond. The microbial flora of a cultivated fish is a reflection of its aqueous environment (Sugita *et al*., 1985; Oni *et al*., 2013).

Fish ponds are constructed where fishes are fed and their growth are easily observed and monitored. In Nigeria, catfish is produced in 98% of our fish ponds. The species of catfish that can be produced include: *Clarias anguillaris, Clarias gariepinus, Heterobranchus longifilis, Heterobranchus bidorsalis,* etc.

Water is very essential in fish pond, water plays a vital role in the proper functioning of earth ecosystem and also essential for fish and living creatures for metabolism. The temperature of water supplied to a fish pond ranges from 25°C to 35°C as this supports the growth of the microorganisms and fishes found in the pond. There are various sources of water, including well water, borehole water, stream water, river water, etc., that can be supplied to the fish pond. Some bacteria coliform groups like *E. coli*, in the ponds are transported from these sources of water or the media of transportation into the ponds. There are several microorganisms found in ponds including bacteria, fungi, algae, protozoa, nematodes and viruses. Bacteria has a unique characteristics, they are ubiquitous in every habitation on earth, growing in soil, acidic hot springs, radioactive wastes, water and the live bodies of plants and animals (Fredrickson, *et al., 2004*). Thus, bacteria are important microorganisms in ponds, whereby, some are beneficial, others are not.

In Nigeria, the rearing of African catfish (*Clarias gariepinus*) is helping in the provision of food and its usefulness cannot be over-emphasized as the demand for fish is expanding rapidly throughout the world (Ugwuba and Chukwuji, 2010). There is an intricate relationship between the fish, their biotic and abiotic environments and changes in one component may reflect and affect the other (Wurt, 2000). Pond water or water used in intensive fish rearing contain many microorganisms which could be pathogenic or opportunistic pathogen to fish, human, and planktons (Zmyslowska *et al*., 2003). These contaminating microorganisms have been attributed to questionable water quality which can be traced to the water sources and high stocking densities (Okpokwasili and Akujobi, 1996). Water sources for earthen ponds are usually untreated surface water runoffs from streams, rivers, lakes, stored waters while underground water source is being utilized for most concrete ponds.

The African catfish (*Clarias gariepinus*) though naturally hardy is still susceptible to parasites and diseases which are caused by pathogenic microbial flora from gastro-intestinal tract (GIT) which may lead to high mortality during production and high rate of economic loss to the farmer (Kabata, 1985) and which if left uncurtailed in some cases cause infection of man and other vertebrates that consume them (Shawn, 1997). Several species of bacteria and fungi were found to be associated with fish species (Lio-Po *et al.*, 1992). Parasites and diseases, which are caused by the presence of pathogenic microbial flora, reduce fish production by affecting the normal physiology of fish (Kabata, 1985) and which, if left uncurtailed, can result in mass mortalities of fish, or in some cases, infection to man and other vertebrates that consume them (Shawn, 1997). The gut flora consists of the micro-organisms that normally live in the digestive tract of animals (O'Hara and Shanahan, 2006).

Against the above background, if fish is going to play a major role in both providing the much needed protein and contributing to the national economy, current information on such aspect as fish borne diseases is required. Also, if aquaculture will reduce the gap between supply and demand for fish and fishery products then there is need to establish the bacterial flora of farmed fish, pond water and of sediment in the ponds and dams. Therefore, this study seeks to investigate the microbiological status and physicochemical parameters of fish and water samples in selected fish farms in South eastern part of Nigeria.

Thus, the main aim of this study is to investigate the microbiological status and physicochemical parameters of fish and water samples in selected fish farms in South East, Nigeria with the following specific objectives which are;

1. To determine the physicochemical parameters of pond water from different sampling farms.
2. To enumerate the microbiological load of fish and water samples from different sampling farms.
3. To determine the hygiene of the fish cultured and its safety for human consumption

Materials and Methods

Study Area

The study was carried out in Awka city, Anambra state, Nigeria were selected fish farms were sampled for laboratory analysis. Awka is made up of two local government areas, namely: Awka South and Awka North. The town situates at latitude of 6.210528^0N and longitude of 7.072277^0E. Most of the farms combine fish production and livestock production, thus they operate integrated fish production having both concrete, plastic and earthen fish ponds stocked with Majorly Catfish which was used for the Experiment.

Collection of Water and Fish Samples

The water and fish samples were collected from four different locations namely, Ebenebe, Amansea, Amaenyi and Okpuno in Anambra State. Water samples were aseptically collected from the ponds weekly using sterile screw capped bottles and transported to Maeuve Academic Research Laboratory for Physico chemical Analysis and Microbial status. Composites samples were obtained by collecting at different sampling points and depths of 30cm below the water surface. The fish samples were also collected weekly and transported to the laboratory in an ice-packed container for microbiological Analysis. The samples were transported to Zidan Medical Diagnostic center for thorough examination.

Equipment's

The equipment's used include, burette, heating mantle, magnetic stirrer and shaker, 250ml and 250 beaker, 250ml conical flask, timer, digital weighing balance, Buchner funnel, Digital thermometer, calibrated sachi, pH meter, pH paper, digital electrical conductivity meter, 50ml BOD bottle, stopper, Whatman filter paper N0:41, heating flask, porcelain disk, microscopic slide, slid cover, counting chamber,conical flask, 250ml and 500ml beaker, inoculating loop,cotton wool, burnsen burner, petri dish and photometer.

Reagents

The reagent used include alkali iodide-azide reagent, H_2SO_4, sodium thiosulphate solution, starch indicator, methyl red indicator ,EDTA salt, NaOH, boric acid, HCL, acetic acid, phenoldisulphonic, KOH, NH4OH, hydrogen peroxide, Lugo iodine, ethanol, crystal violet, acetone, Nutrient Agar, kovac acid , glucose, bromo thymol indicator, Agar, Nutrient Gelatin, tryptone, MaCconkey agar and Erosin Methylene blue.

Physio-chemical Analysis of Water Samples

The physicochemical parameter of water checked include, temperature, Electrical conductivity, total dissolved oxygen, total dissolved solid, pH, Water hardness, and Turbidity. The water samples from the different fish farms were examined for physiochemical characteristics using standard procedures of the American Public Health Association (APHA). pH was measured in-situ using a pH meter after it has been calibrated with standard buffer7, pH paper was also used to confirm the reading as well. Temperature and conductivity were measured using a digital thermometer and electrical conductivity meter. The Modified winkler's method was adopted for dissolved oxygen determination, Gravimetric method was used for Total Dissolved Solid, Titrimetric method was used to calculate Water Hardness, Turbidity was measured using a hygrometer disc and Biological oxygen demand was calculated using the Iodometric method.

Microbial Isolates from Pond Water

Preparation of Culture Media

One gram of nutrient agar, MaCconkay agar and Erosin methylene blue agar were dissolved in 45ml of distil water, then autoclaved for 15min under the pressure of 15 pounds' pressure(psi) at a temperature of 121^0C. The media were poured into petri dish and allowed to cool into cake form ready for inoculation. The samples were inoculated into the culture media then incubated for d 24-48hours for the growth of bacteria. After the incubation period, the observed growth was subculture to get a pure culture.

Bacteria Colony Load Count: Direct colony count and microscopic method by W.H.O (2010) was used. In this method, the colonies of bacteria were counted directly from the cultured plate and further countered under microscope.

Bacteria Biochemical Test and Identification

Coagulase test: three percent (3%) hydrogen peroxide was drop on a smear of bacteria on a slide, the liberation of oxygen gas a positive test for bacteria capable of producing catalase enzymes.

Oxidase test: Heavy lump of bacteria inoculum was introduce into one percent glucose in the presence of brom thymol blue or bromocerol blue indicator, then incubated for 24-48 hours, the decolourization of the solution into green or yellow is an indication of the ability of the bacteria to cause oxidization as well confirm the presence of *Aeromonas spp*

Motility test: smear of bacteria was made on a slide with drop of distil water added and view under the magnification 100 to ascertain the motile ability of the bacteria

Indole test (Kovac test): light inoculum of the bacteria was inoculated into tryptone broth media and incubated for 24hours. Light inoculum of the bacteria was isolated from the media and introduced into sterile test tube. Five drops of kovac reagent was added to the tube and agitate the appearance of cherry red ring form at the surface of the media confirms the presence of *E.coli.*

Glucose fermentation test: To one percent (1%) brom thymol blue indicator was added and 10µl of distil water was added. A sterile needle was use to introduce a colony of bacteria into the solution then incubated for 24-48 hours for acid or gas production the production of gas and as well as the decolourization from green to yellow is an indication that the bacteria is capable of causing oxidation and indicates the presence of aerobic organism.

Alternative test for Gram Reaction: A drop of three percent (3%) solution of KOH was drop on a slide and a lump of bacteria drop on it. Gram positive bacteria change the viscosity of KOH solution while gram negative bacteria makes KOH solution to become stringy or mucoid in appearance and consistent.

Gram Staining Test: A drop of water was made on a slide and an isolate colony of bacteria smeared on the slide, mix with the d droplet of water on the slide till becomes turbid. It air dried and then heat fixed at very low temperature and allowed to coll. The slide was flood with crystal violet solution and allowed to stand for 60seconds and then washed off. Gram's iodine was used to flood the smear, allowed for another 60 seconds and washed off with water. It was then decolourized with 95% alcohol and 5% acetone solution till the solvent flows colorless from the slide. The slide with the fixed organism was rinsed in water the counter stain with safranin for 60 seconds, rinse and allowed to air dry then viewed under microscope. Gram negative bacterial take up a pink or red colour of the counter safrannin while gram positive retain the crystal violet and remain purple to dark blue colour.

Fish Sample Collection and Culturing

A small portion of each of the grinded fish sample weighing 1 g was transferred aseptically into 9 ml of distilled water in a test tube. Samples were serially diluted into 4-fold using 1:10 dilution. 1 ml of the dilution was taken from the 4^{th} test tube into two sterile petri dishes with labelling. Nutrient agar was added into the petri dishes accordingly, mixed and allowed to gel together. The nutrient agar was then incubated at 37^{o}c for 24 hours.

Plate Reading and Bacterial Isolation from Fish Samples

After 24-hour incubation, bacterial growth on nutrient agar was observed and counted. Different bacterial colony was isolated and sub-cultured according to their colony morphology.

Statistical analysis

Results collected were subjected to analysis of variance and the least significant difference (LSD) test was used to separate differences between means at 5% probability level.

Results

The mean values of the Physico chemical parameters of the water from Ebenebe, Amansea, Amaenyi and Okpuno are presented in table 4.1. There was a significant difference ($p<0.05$) in all the physical and chemical parameters of the water from all the locations except in the temperature and pH of which $p>0.05$. The overall results of water samples in the four locations showed variations in the distribution of their range (mean) such that the level of pH was highest im Amaenyi (6.98 ± 0.05) and lowest at Okpuno (6.00±0.02), conductivity was highest in Okpuno (0.08±0.02) and lowest in Amansea (0.02±0.02), in addition, TDS was highest at Ebenebe (0.25±0.13) and lowest in Okpuno (0.02±0.02), while D>O was highest at Amansea (5.50±0.48) and lowest value was observed at Okpuno (4.68±0.29).

However, the level of BOD was highest at Okpuno (3.88±0.28) and lowest at Amansea (2.75±0.50), temperature was highest at Amansea (26.07±0.50) and lowest value was observed at Amaenyi (25.5±0.16). additionally, water hardness level was highest at Amaenyi (0.95±0.06) and lowest at Okpuno (0.61±0.50) while turbidity level was highest at Amansea (5.42±0.59) and lowest at Okpuno (2.50±0.29).

Table 1. The Mean values of Physicochemical parameters in pond water from Ebenebe, Amansea, Amaenyi and Okpuno.

Parameters	**Ebenebe**	**Amansea**	**Amaenyi**	**Okpuno**	**WHO/NSDWQ**
pH	6.75 ± 0.50 [a]	6.50 ± 0.58 [a]	6.98 ± 0.05 [a]	6.00 ± 0.82 [aaa]	6.5-8.5
Conductivity(amp)	0.03 ± 0.01 [a]	0.02 ± 0.02 [b]	0.03 ± 0.01 [c]	0.08 ± 0.02 [abc]	500/1000cm
TDS (ppm)	0.25 ± 0.13 [a]	0.04 ± 0.00 [b]	0.25 ± 0.06 [c]	0.02 ± 0.02 [abc]	1000mg/l
D.O (mg/l)	5.15 ± 0.13 [a]	5.50 ± 0.48 [b]	4.75 ± 0.24 [c]	4.68 ± 0.29 [abc]	5-14mg/ls
BOD(mg/l)	2.77 ± 0.59 [c]	2.75 ± 0.50 [d]	2.75 ± 0.65 [e]	3.88 ± 0.28 [cde]	6mg/l
Temperature (^{0}c)	25.75 ± 0.89 [a]	26.07 ± 0.50 [a]	25.50 ± 0.16 [a]	26.05 ± 0.82[aaa]	31-34^{0}c
Water Hardness	0.80 ± 0.19 [a]	0.76 ± 0.14 [b]	0.95 ± 0.06 [c]	0.61 ± 0.06 [abc]	75mg/l
Turbidity(NTU)	3.50 ± 0.59 [a]	5.42 ±0.57 [b]	4.40 ± 0.18 [c]	2.50 ± 0.29 [abc]	10Ntus

Means with same superscripts on the same row are not significantly different ($p>0.05$).

The result of the bacteria load in water sample from the four sampled stations are represented in table 4.2; *Salmonella spp* and *Staph. Spp* had the highest and lowest bacterial count respectively in Ebenebe pond water while in pond water from Amansea, *vibrio spp* has the highest bacterial count but showed least/no bacterial count for *staph spp* and *acinetobactercalcoaceticu.* The pond water gotten from Amaenyi showed the highest bacterial count for salmonella spp but showed least/no bacterial count in *Plesiomonas spp, acinetobactercalcoaceticu* and *Yersinia spp.* Furthermore, *Plesiomonas spp* and *Salmonella spp* showed the highest bacterial count in Okpuno water but *Yersinia spp* and *Staph. Spp* showed least/no bacterial count.

Table 2: The Mean values of Bacteria isolates from Ebenebe, Amansea, Amaenyi and Okpuno pond water.

Bacteria	**Ebenebe**	**Amansea**	**Amaenyi**	**Okpuno**
Plesiomonas spp	2.50±0.70	1.00±0.00	0.00±0.00	3.00±0.00
Escherichia spp	2.50±0.71	2.50±0.72	2.00±1.41	2.00±0.00

Staph. Spp	1.00±0.00	0.00±0.00	2.00±0.00	0.00±0.00
Pseudomonas	2.00±1.41	2.50±0.71	1.50±0.71	1.00±0.00
Vibrio spp	2.50±0.71	3.00±0.00	2.00±1.41	1.50±0.71
***A** Bactercalcoaceticu*	1.00±0.00	0.00±0.00	0.00±0.00	2.00±0.00
Salmonella spp	3.00±0.00	3.00±0.00	3.00±0.00	3.00±0.00
Yersinia spp	2.00±0.00	1.50±0.71	0.00±0.00	0.00±0.00
Plesiomonas spp	2.00±0.00	1.50±0.71	1.50±0.71	2.00±0.00

Table 3 The Bacterial Isolates Identified Using Biochemical Test

Bacteria organisms	**Indole test**	**Motility test**	**Glucose test**	**Oxidase test**	**Coagulase test**	**Grams staining Test**
Plesiomonasspp	-	-	-	-	-	-
Escherichia spp	+	+	+	+	+	Gram negative
Staph. Spp	+	+	+	+	+	Gram positive
Pseudomonas spp	-	-	-	-	-	-
Vibrio spp	-	-	-	-	-	Gram negative
Acinetobactercalcoaceticu	-	-	-	-	-	-
Salmonella spp	+	+	-	+	+	Gram negative
Yersinia spp	-	-	-	-	-	-

Table 3: shows the results of some bacterial isolates identified using biochemical test like Indole test, Gram staining test, Oxidase test et cetera.

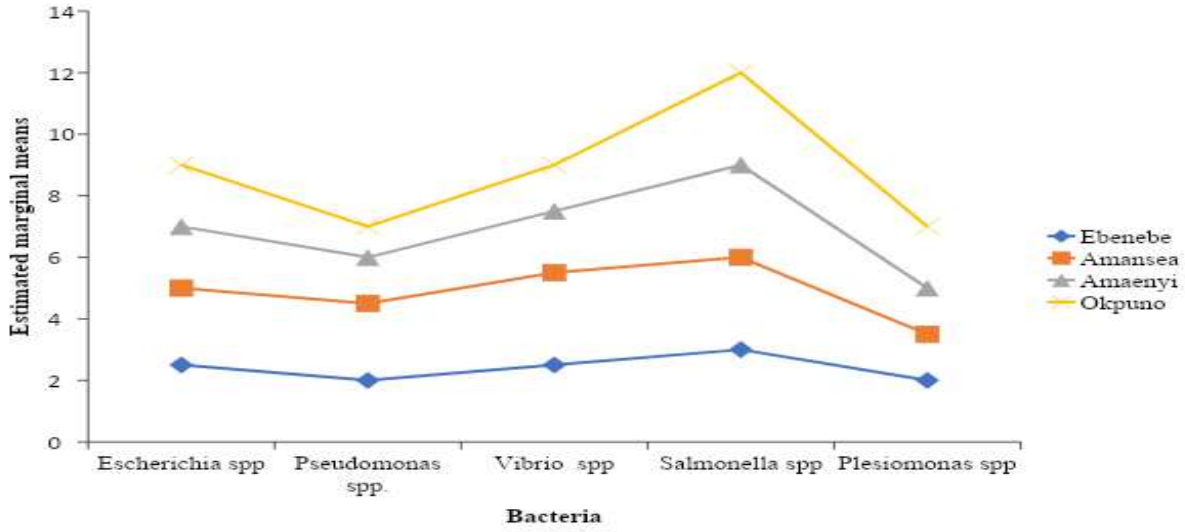

Fig 1: Estimated Means of Observation

Figure 1 shows the estimated means of observation of bacteria isolated from the sampled farms. *Eschericiaspp, pseudomonas spp, Vibrio spp and Salmonella spp and plesiomonas spp* in Ebenebe, Amansea, Amaenyi and Okpuno pond water.

Table4 Table showing the Bacterial Load in Fish.

LOCATION	**Bacterial Load X10^5cfu/G**
A	105.00±36.86
B	79.00±16.52
C	108.33±11.06
D	97.66±18.33

A= Ebenebe, B = Amansea, C = Amaenyi & D = Okpuno

The result shows that the highest bacteria load was observed in fishes from station C while the lowest was at station B.

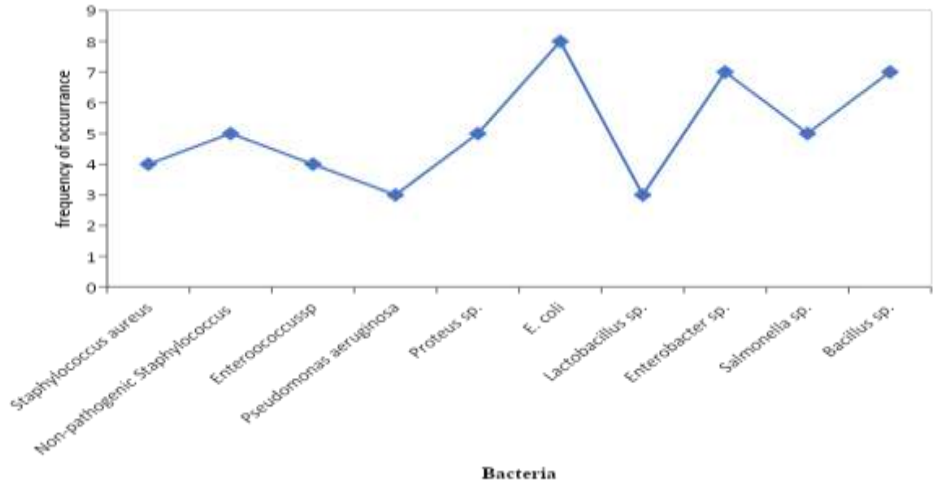

Fig 2: Table showing the Frequency of Bacteria Occurrence in Fish

Figure 2 shows the frequency of Bacteria Occurrence In fish. *E. coli* showed the highest frequency in fishes samples, followed by *enterobacter spp* and bacillus spp. *Lactobacillus spp* showed the list frequency followed by *pseudomonas aeruginosa*.

Table 5: Biochemical Test for Bacteria in Fish

Isolates	**Gram staning**	**Indole**	**Urease**	**Cit**	**CA**	**MR**	**CO**	**SSA**	**Suspected bacteria**
1	+ cocci	-	-	+	+	+	-	-	*Non-pathogenic Staphylococcus*
2	+cocci	-	-	+	+	-	+	-	*Staphylococcus aureus*
3	+cocci	-	-	-	-	-	-	-	*Enterococcus sp.*
4	-rod	-	-	+	-	+	-	-	*Pseudomonas sp.*
5	-rod	+	-	-	-	+	-	-	*E. coli*
6	-rod	-	+	+	+	+	-	-	*Proteus sp*
7	+rod	-	-	-	+	-	-	-	*Lactobacillus sp.*
8	+rod	-	-	+	+	-	-	-	*Bacillus sp*
9	-rod	-	-	-	+	+	-	+	*Salmonella sp.*
10	-rod	-	-	+	-	+	-	-	*Enterobacter*

Discussion

The present study elucidated results of physico-chemical parameters in pond water from Ebenebe, Amansea, Amaenyi and Okpuno in Anambra state Nigeria between April to June, 2021. In this study the pH of water gotten from Ebenebe, Amansea, Amaenyi and Okpuno was fluctuated between the range of 6.0 – 6.9 where Okpuno was the least (6.0) and others had pH ranging from 6.5 – 5.9 but there was no significant difference among the pond waters from the four stations. The pH obtained in this study is in line with the limit acceptable by Food and Agricultural Organisation standard (FAO 2009) and WHO standard (W.H.O 2006) maximum containment level of 6.5-9.5 and was similar to that of Ehiagbonare and Ogunrinde (2010) who studied the physiochemical analysis of fish pond water in Okada in Edo state. Ntengwe and Edema (2008) observed that the appropriate pH for increased fish production is 6-9.

The electrical conductivity of Ebenebe, Amansea, Amaenyi and Okpuno where 0.035 ± 0.01, 0.025 ± 0.02, 0.03 ± 0.01 and 0.08 ± 0.02 respectively, although there was a significant difference between them ($p<0.05$) Table 4.1. The conductivity values obtained from this study falls within the FAO, (FAO, 2009) and WHO (W.H.O, 2006) limits of 1000 µS/cm(0.1amp), and differs with the report of (Babatunde and Woke, 2015) who observed higher values of 78 µS/cm – 1428.50 µS/cm(0.0078-0.1428amp) from physicochemical burden of Oyo State fish pond, Ibadan, South-West Nigeria, so the studied fish pond waters would be regarded as safe for fish production. Total Dissolved Solids (TDS) is an indication of the amount of dissolved substances (Umeh *et al,* 2020). Total dissolved solids values obtained from the study ranges from 0.02-0.25ppm of which there was a significant difference between the four-pond water. The finding of this study is in line with acceptable limit for TDS (1000mg/l) set by FAO (FAO 2009) and WHO (W.H.O, 2006).

Low concentration of dissolved oxygen (DO) in water causes suffocation in fish while its super saturation may result into the gas bubble disease leading to the mass mortality of fish in both cases. Furthermore, the solubility of oxygen in water decreases as the water temperature increases (Umeh *et al.,* 2020). FAO and WHO acceptable limit for dissolved oxygen stipulates that is must be range between 5-14mg/dl to be safe which didn't agree totally with the result of this study. Ebenebe and Amansea water showed DO levels >5mg/dl while Amaenyi and Okpuno showed DO <5mg/dl which is quite lower than the recommended value of WHO and therefore not suitable for high fish productivity. Similar results were also reported by Okpokwasilli and Ubah (1991), Who worked on water quality and bacteria disease in fish ponds.

The BOD level obtained in this study ranges from 2.75mg/dl to 3.88mg/dl which is within FAO and W.H.O (2009) value of 6mg/l. These BOD values are therefore within the values for optimum fish activities. The values were similar to the values (2.98 mg/l to 4.27 mg/l) reported by Umeh *et al.* (2020). Fish is a poikilothermic animal, so its temperature is dependent on the temperature of its environment. It changes with the temperature of the surroundings and the temperature values observed in the fish ponds ranged from 25.08^0C to 26.08°C (Table 4.1) which is within FAO and W.H.O (2009) maximum containment level goal (25°C-32°C) but the samples was not significantly different ($p>0.05$). Thus, this corroborates with the report of Fafioye (2011), who observed a temperature of $27\text{-}28^0$C.

The water hardness observed in the fish pond water samples ranged from 0.61mg/l to 0.81mg/l (Table 4.1). The values are in line with the FAO and W.H.O (2009) permissible limit of 75 mg/l and therefore suitable for fish farming. The values were similar to the values (6.19 mg/l to 37.58 mg/l) reported by Torimiro *et al.* (2014). Turbidity values reported in the fish pond waters ranged from 2.50 NTU to 5.42NTU (Table 4.1) and were within the FAO and W.H.O. (2009) permissible limit of 10 NTU. The turbidity results were similar to the values (7.73 NTU to 11.33 NTU) recorded by Agbaire *et al.* (2015). However, there was a significant difference between the sample pond waters ($p<0.05$).

Bacteriological analysis of the water samples showed nine different genera (table 4.2) which are *Plesiomonas spp, Escherichia spp, Staph. Spp, Pseudomonas spp, Vibrio* spp, *Acinetobactercalcoaceticu,*

Salmonella spp, Yersinia spp and *Plesiomonas spp.* The water obtained from Ebenebe fish pond showed the highest number of bacterial load having the nine different genera present in it while the Amaenyi pond water showed the least level of bacterial contamination having *Plesiomonasspp, Escherichia spp, Pseudomonas spp, Vibrio* spp, *Salmonella spp* and *Plesiomonas spp. Salmonella spp* showed the highest bacterial load of 3.0 x 10^5cfu/g in Ebenebe water admist other bacteria while *Acinetobactercalcoaceticu* was observed to have the least bacterial count of 1 x 10^5cfu/g as supported by Umeh *et al,* (2020).

Salmonella spp causes salmonellosis, which in humans could result in severe typhoid fever (enteric fever), or salmonella fever and bacteremia (Egbere *et al.,* 2010). Likewise, *Salmonella spp* and *Vibrio* spp showed bacterial load in Amansea pond water of 3.00 x 10^5 and 3.00 x 10^5cfu/g respectively while *Plesiomonas spp* was discovered to have the least bacterial load of 1.00 x 10^5cfu/g. This finding is similar with the findings of Njoku *et al.* (2015) where the bacterial load of salmonella was in concrete pond was 3.2 x 10^5 cfu/ml and vibrio spp was 5.9 x 10^4cfu/ml. *Pseudomonas spp* especially *aeruginosa* could cause general inflammation and sepsis in critical body organs such as lungs, kidneys, urinary tract, which can be fatal because it thrives in most, surfaces (Udeze *et al.,* 2012).

The result also revealed that *Salmonella spp* had the highest bacterial load in both Amaenyi and Okpuno pond water, *Escherichia spp* and *Yersinia spp* had a maximum bacterial load of 2.50± 0.72x 10^5cfu/g and 2.00±0.00x 10^5cfu/g respectively. The presence of pathogenic microorganism especially *E. coli, salmonella, shigella and Vibrio* can lead to the transmission of water borne diseases such as typhoid fever, cholera, food poisoning and gastroentritis (Piet, 2009) on consumption of improperly cooked fish cultureed in theses ponds or through contact with the contaminated fish and water. The biochemical test result shows that 75% of the bacteria present are gram-ve.

In this study, there was no significant difference ($p>0.05$) in the total bacterial load in fish gotten from A, B, C and D. Although, C showed the highest total bacterial load of 108.33±11.06x10^4cfu/g followed by A with bacterial load of 105.00±36.86x 10^4cfu/g and B having the least total bacterial load of 79.00±16.52x 10^4cfu/g. Moreover, *E. coli* showed the highest frequency of occurrence in fishes' samples, followed by *enterobacter spp* and bacillus spp. *Lactobacillus spp* showed the least frequency followed by *pseudomonas aeruginosa*. This result is similar to that of Ogeneogaga and Solomon, (2017) where *E. coli* had the highest frequency of occurrence (25%) in fish samples.

In general, the finding of this study in relation to bacteria load in fish from the sampling stations it was observed that the bacteria loads of fish is in line with the acceptable limit set by International Commission on Microbiological Specification for Food, Fish and Fishery Products (ICMSF, 1986) who opined that any fish that contain more than x 10^6 bacteria count is not suitable for human consumption.

Conclusion

The present study revealed that pond waters involved was contaminated by bacteria, which might affect the growth of fish in the pond and may become harmful to human due accumulation of these bacteria in fishes and also in food-chain over time. The difference in physicochemical properties of the pond water was significant but was still within safe limits stipulated by FAO and WHO which makes them suitable for fish rearing. Although there was variation in concentration of bacterial load but there was no significant among the sample ponds. The presence of *Salmonella spp* in all the pond water of which it ranked highest in all the water sample portrays danger although it was still within safe limits. Therefore, bacterial population the sample water ponds where similar but the physicochemical properties significantly different which might be attributed to the location of the sample pond waters. Also, the bacteria load of the fish was observed to be within the acceptable limit making the fishes suitable for human consumption.

Recommendation

From the result gotten from the study, it is therefore recommended;

1. That Fishes should be properly cooked before being consumed to limit the bacteria from infecting humans.
2. That microbiological analysis and physicochemical examination of wastewater or fish for signs of possible infections should be conducted on a regular basis.
3. That the ministry of agriculture should ensure that the fish farmers are supplied with healthy fry for their stock.
4. That Good water quality such as well or borehole should be used in the fish pond rather than water from questionable sources such as river, stream, and surface runoff.
5. That the fish feeds should be sourced from reputable manufacturers, Water in the fish pond should be changed completely at regular intervals.
6. That Waste water should be treated either by physical methods which involve filtration through slow sand filters, rapid sand filters, sand beds or chemical methods such as addition of disinfectants for example chlorine before final disposal into surrounding drains.

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Prevalence of Zoonotic Tuberculosis in Cameroon's Wildlife

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Abstract
This study was carried out between March to December 2022 in the south and east regions of Cameroon. Veterinary inspections were carried out on wild animal carcases, in "bush markets". Two hundred and ninety two (292) carcasses of, antelope 109 (37.33%), doe 26 (8.9%), porcupine 72(24.66%), civet 5(1.79%), wild boar 3(1.3%) and monkey 16 (5.48%), tiger cat 4 (1.37%), civet cat 5(1.71%), Hare 15(5.14%), pangolin 32(10.96%), Rat 9(3.08%), varan 1(0.34%), were sampled for this study. Lungs, livers, spleens and lymph nodes were harvested from these animals and subjected to standard laboratory test for isolation and identification of Mycobacteria. The result shows that Culture on Lowenstein Jensen solid medium yielded a prevalence of 115 (39.38%) colonies of mycobacteria from all samples collected; observation of BAARs by ZN staining revealed 34 (11.64%) positive samples for mycobacteria from all samples. Biochemical test positivity gave 56 (19.18%) positive samples. Mycobacteria identifiable on the basis of morphological characteristics were *M. intracellulare*, *M. xenopi* and *M. gordonae*. The real-time PCR revealed (0%) mycobacteria of the *Mycobacterium tuberculosis* complex. This study showed that mycobacteria of the *Mycobacterium tuberculosis* complex were not detected in wild animal but atypical mycobacteria were found. Wildlife during our study period cannot be considered as a possible reservoir of mycobacteria of CMT, given the non-proximity with cattle breeding sites and the rarity or absence of contact between wildlife and cattle.

Keywords: Tuberculosis; Prevalence; Mycobacteria; *Mycobacterium Tuberculosis* complex; Wildlife; South and East; Cameroon

Introduction

Animal tuberculosis is a global disease caused by bacteria belonging to the *Mycobacterium Tuberculosis* Complex (MTC). It can infect humans and a wide range of domestic and wild mammals (Caminiti et al., 2016). Several species of mycobacteria exist including *M. tuberculosis*, *M. bovis, M. africanum, M. caprae*, *M. microti*, all belonging to the *M. tuberculosis* complex (CMT) (Awah, 2012). Highly prevalent worldwide, *M. tuberculosis* and *M. bovis*, are the main agents responsible for zoonotic tuberculosis with serious risks for human health worldwide, with an estimated 9.9 million people having developed tuberculosis worldwide according to WHO reports in 2021 (Serges et al., 2021). Animal tuberculosis is of threefold interest: firstly, in economic terms, due to the costs associated with diagnostic tests and the slaughter of infected livestock; secondly, in terms of hygiene, due to the interaction between animals and

humans (Zanelle, 2007) and thirdly, in medical terms, due to the mortalities it causes. Despite all the efforts made to eradicate the disease, there still seems to be a considerable amount of carriage. In some countries, the failure to eradicate bovine tuberculosis, despite the implementation of control measures, has been attributed to the presence of the disease in wildlife. Indeed, in these countries, bovine tuberculosis appeared secondarily in wild species, some of which became reservoirs (common warthog, African buffalo, lechwe antelope, african elephant all in South Africa and Eurasian wild boar in North Africa (Zanelle, 2007). As a result, *M. bovis* in wildlife is as much a public health issue as bovine tuberculosis in cattle, this disease is endemic in African countries and Cameroon is not an exception. Several studies carried out in Cameroon have shown the endemicity of tuberculosis in cattle, characterizing *M. bovis* tuberculosis as the fifth zoonosis among the priority zoonotic diseases in Cameroon (Awah et al., 2012). According to work carried out by Awah et al., (2012) its average prevalence is 5.53% (3.59- 7.48) in cattle reared in the highlands of Cameroon. This endemic status of *M.bovis* tuberculosis in cattle remains unresolved in Cameroon's wildlife. In order to remedy this deficiency, this study was initiated to determine CMT tuberculosis in the wild animal population of the South and East regions of Cameroon. This study is a first in Cameroonian wildlife.

Materials and Methods

Study Areas

The regions of South located between 2° 30′ north, 11° 45′ east, over an area of 47,110 km2 and East located between 4° north, 14° east, extended over an area of 109,002 km. These regions are forested areas with dense, diverse vegetation, conducive to the development of agricultural activities and wildlife. Because of the diversity of wildlife (Samson, 2001). Hunting is the main source of protein for rural communities and one of their main sources of income, due to the abundance of game in these areas.

Materials

Our study was a descriptive cross-sectional study. It took place from March to December 2022. The biological materials were lung, liver, spleen and lymph nodes with or without suspicious lesions of tuberculosis. Samples were collected in sterile bags and transported to the laboratory (LANAVET) in a cooler containing carboglass, for storage at -20 degrees Celsius. The samples were then processed and analyzed for mycobacteria isolation. Culture on Lowenstein-Jensen solid medium, to detect mycobacteria; Ziehl-Neelsen staining for the detection of bacilli alcaloo acido resistants (BAARs); biochemical tests such as urease, catalase and nitrate reductase for the detection of mycobacteria; and molecular analysis by real-time PCR for the detection of mycobacteria of the *Mycobacterium tuberculosis* complex (MTC).

Study Population

The present study was carried out on wild animal corpses (antelope, doe, civet, baboon, wild boar, porcupine, pangolin, and rat). All wild animal corpses putrefied or not, were included in our study. For sampling, we used (Survey Toolbox Ausvet, 2015), N = Z2. P (1-P) /m 2

N= Sample size; Z= Confidence level according to the normal distribution, Z= 1.96 if the confidence level is 95% (Standard deviation value of 1.96); m = margin of error at 5%.

We have considered a prevalence of 18.26% (obtained from MINEPIA) in 2016. The total number of animals to be sampled was around 230.

Statistical Analysis

The data collected were recorded in a Microsoft Excel 2010 spreadsheet program and analyzed using R software version 4.1.0. A descriptive analysis to determine frequencies was performed.

Results

The different families of species encountered: Family Bovidae with the antelope (*Cephalus dorsalis*) as species. Family Cervidae with the doe (*Cervus elaphus*) as species. Family Felidae with the tiger cat

(*Leopardus wiedii*) as species. Family Virerridae with the civet cat (*Civettictis civetta*) as species. Family Manidae, including the pangolin (*Phataginus tetradactyla*). Family Arteridae, with the porcupine (*Arterus africanus*) as species. Family Nesomyidae, with the rat (*Cricetomys emini*) as species. Family Suidae with the wild boar (*Sus scrofa*) as species. Family Varanidae with the *Varanus varius* as species. Cephalophe family, including the hare (*Cephalophe bleue*). Cercopithecidae family, including the baboon (*Papio anudi*)

Table I: Distribution of species sampled

	N	%
Antelope	109	37.33
Doe	26	8.9
Tiger cat	4	1.37
Civet cat	5	1.71
Hare	15	5.14
Pangolin	32	10.96
Porcupine	72	24.66
Rat	9	3.08
Wild boar	3	1.03
Baboon	16	5.48
Varan	1	0.34
Total	292	100

Table I shows that the wild animal mostly found was the antelope (*Cephalus dorsalis*), 109 (37.33%) from the family of *Bovidae*.

Table II: Frequency of growths on Lowenstein-Jensen solid medium following inoculation with samples

	N	Proportion for culture (%)
Negative	177	60.62
Positive	115	39.38
Total	292	100

This shows that 115 (39.38%) of the inoculated samples produced colonies.

Table III: Frequency of ZN (Ziehl-Neelsen) stained BAARs on organs from wild animals in the South and East regions of Cameroon

	N	Proportion coloration (%)
Negative	258	88.36
Positive	34	11.64
Total	292	100

ZN staining revealed the presence of

BAARs in 34 (11.64%) samples.

Table IV: Frequency of identification tests on wild animal organs in the South and East regions

	N	Percentage (%)
Negative	236	80.82
Positive	56	19.18
Total	292	100

The catalase test at 22 degrees Celsius, catalase test at 68 degrees Celsius, urease test, and nitrate reductase test were positive on 56 (19.18%) samples.

Table V: Prevalence of mycobacteria of the *Mycobacterium tuberculosis* complex by PCR on wild animal organs in the South and East regions

	N	%
Negative	292	100
Positive	0	0
Total	292	100

Table V shows that no mycobacteria (0%) of the *Mycobacterium tuberculosis* complex were detected.

Discussion

This study is the first to investigate tuberculosis in wildlife in Cameroon. In the south and east regions of Cameroon, several wild species are prized for both personal consumption and marketing, in varying quantities. Antelope (n=109/292, 37.33%) was the most frequently encountered in this study. Its abundance, and at the same time the abundance of organs harvested (lung), may be justified by its accessibility for marketing. This variability in quantity could also be explained by the fact that the abundance of a species in a hunting area varies according to the season and the preferences of the consumers. The predominance of lung among the organs harvested is justified by the fact that lung is not

the organ most consumed by the population. After seeding on solid LJ culture medium, we obtained 115 (39.38%) colonies that grew. This relatively low rate could be explained by contamination of the cultures by spore-forming bacteria. ZN staining revealed the presence of BAARs in 34 (11.64%) samples. This prevalence is similar to that of Tschopp et al. (2010) in Ethiopia, who obtained 29(32.5%) out of 89 wild animals. This result is contrary to that of Angela et al. (2017), in whom no BAARs were observed on mongoose organ samples. Furthermore, the positivity of 56 (19.18%) to biochemical tests (catalase, urease, nitrate reductase) also indicates the presence of mycobacteria. This result is similar to that of Tschopp et al. (2010) in Ethiopia, who detected atypical mycobacteria, in their case *Mycobacterium terrae*. The result is also similar to that of Katale et al (2014) from Tanzania, who isolated by culture, multiplex PCR and spoligotyping fifty-five (55) atypical Mycobacteria (*Mycobacterium intracellulare*, *M. lentiflavum*, *M. fortuitum*, and M. *chelonae-abscessus group*) representing 16 mycobacterial species. These results could be explained by the fact that atypical mycobacteria are present.

The aim being to determine the prevalence of CMT tuberculosis in wildlife, we obtained a prevalence of 0%, through PCR. This result is similar to that of Tschopp et al. (2010) from Ethiopia, who did not detect CMT mycobacteria in wildlife. The 0% true prevalence of CMT tuberculosis detected by real-time PCR in wildlife reported in our study could be explained by infrequent contact between wild and domestic animals, especially infected cattle. Our results differ from those of Hang Ombre et al. (2012) who through PCR identified 15 to 37 isolates of *M. bovis* from lechwe antelope and cattle, in Zambia. Similar results to those of Rosen et al. (2017) in Mozambique, who detected *Mycobacterium tuberculosis* in 35(17.1%) elephants using the tuberculin test.

Conclusion

Based on the prevalence obtained in wildlife using culture, ZN staining and identification tests, we can conclude that infection due to atypical mycobacteria are a pathological entity to be considered in wild animals. However, the remoteness of cattle-breeding sites from our study areas and the scarcity of contact between wildlife and cattle could justify the absence of CMT mycobacteria on PCR. We can therefore say that during our study period, wildlife did not constitute a potential reservoir of zoonotic tuberculosis in Cameroon, as the prevalence of CMT mycobacteria was 0%. This hypothesis undoubtedly constitutes a basis for research that would provide a better understanding of the epidemiology of *M. bovis* tuberculosis in wildlife, a condition that is still poorly understood in our regions.

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Review Update on the Situation of Tuberculosis on the Interface of Domestic Cattle-wild Life in Africa

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Abstract
Bovine tuberculosis eradication is an actively ongoing research subject, mostly in developed countries due to its impact on economy, public health and conservation. Research on bTB in under developed countries still remain stagnant or not really progressing even though it is an endemic disease in cattle. Cattle is considered as the main host of this disease but other studies have shown that several wildlife species are involved in the maintenance and transmission of bTB. Recent studies on the status of bTB at the wildlife-cattle interface have gained relevance in recent years. Awareness on the status of bTB in the African wildlife is a paramount step for ensuring control, surveillance in cattle. This review systematically addresses data available on bovine tuberculosis in Africa's wildlife. This article has given an overview of the factors related to prevalence, host, causal agents, methods of diagnostics, risk factors. After several screenings, six articles were considered for this systematic review. East and Southern Africa countries carry on most researches on bovine tuberculosis in wildlife. Available studies assessed the status of wild mammal, carnivore. Further research to improve the knowledge on the status of bovine tuberculosis in wildlife in Africa are still needed in order to allow proper disease monitoring in wildlife-cattle and human.
Keywords: bovine tuberculosis, interface wildlife- cattle, Africa

Introduction

Tuberculosis is a chronic infectious disease caused by members of the *Mycobacterium* tuberculosis complex (MTBC). It affects livestock, wildlife, and humans (Poirier et al., 2019). The members of this complex have shown the ability to infect a large diversity of natural populations of non-domesticated, and domesticated species worldwide (Caminiti, 2020). Proven to be zoonotic pathogens in public health, it has also been identified in several wild animals including cervids in North America, badgers in Great Britain, feral pigs in Europe, brushtailed possums in New Zealand, and buffalo in South Africa (Fitzgerald and Kaneene, 2012). Bovine tuberculosis becomes detrimental to biodiversity conservation, where it threatens wildlife populations or when disease management options consider the possibility of wildlife control (Alexandre Caron et al, 2014). In Africa it is a regional threat; the epidemiological situation of bTB in livestock and human populations is highly variable (Hutcheon, 1880), reasons being there are very few countries with a structured surveillance system to know its distribution, and information about its occurrence. The first recorded case of bTB in Africa's cattle was diagnosed in South Africa in 1880. Although bTB in cattle occurs widely in Africa, the diagnosis is mainly dependent on the detection of macroscopic lesions during

meat inspection of carcasses in abattoirs (Awah-Ndukum et al. 2012; Egbe et al. 2016). In Africa's wildlife the first detection of *M.bovis*, was during a destructive outbreak of the disease in South Africa (Paine and Martinaglia 1929) in greater kudus (*Tragelaphus strepsiceros*) and a few other wildlife species. Several studies in Sub Saharan Africa have shown the existence of tuberculosis in cattle and wildlife. Though knowledge gaps remains, the spillover of bTB from cattle to wildlife has already been described on several occasions; affecting a diversity of wildlife species in a wide range of protected areas of Sub-Saharan Africa. (Jori et al. 2013). This study aims to review the present state of bovine tuberculosis in Africa's wildlife, in other to see how far the gaps on the awareness has been filled.

2. Method

A systematic bibliographic review was conducted on the situation of bovine tuberculosis in Africa's wildlife; by searching PubMed, Google Scholar and Agora. No time limitation was imposed. The search criteria were specified in advance and the search was executed on 31/03/2022 and last updated on 26/03/2023. The search string used was the following: "endemics'' OR "epidemiology'' OR "risk factors'' OR "population at risk'' OR "health correlates'' AND "bovine tuberculosis'' OR "mycobacterium bovis infection'' AND "cattle'' AND "wildlife'' AND "Africa''. Abstracts selection was according to their relevancy and excluded if dealing exclusively with cattle, or human, or other geographical regions out of Africa. Articles were reviewed in full text. For each article, the extracted data included year of publication, authors, country of the study, type of study, method of study, number of animal, number of positive sample, wildlife host, and risk factors. Information were summarized and presented in table format.

3. Results

The bibliographic search yielded 171 articles. These searches were combined and duplicates were removed. Fig 1 depicts the flow of the selection process. One hundred and fifty-eight (158) articles were excluded after screening. One hundred and thirty-one (131) articles were assessed for eligibility, fourteen (14) were excluded with reasons that didn't meet all inclusion criteria. Six (6) articles were selected as of interest to the present review.

3.1- Characterization of Published Articles

The studies were published between 2002-2023. The regional distribution of studies was East Africa (4), Southern Africa (2). The studies enrolled six authors from five countries. The regional distribution of studies was Mozambic (1), Zambia (1), Tanzania (2), South Africa (1), Zimbabwe (1). The follow up duration of the study ranged from three to seven years. The studies were of high quality. Types of studies were cross sectional, and retrospective studies. Most of the studies were conducted on the field. Approximatively all the included studies were published during the last decade of our review as from 2010 to 2023 in table 1.

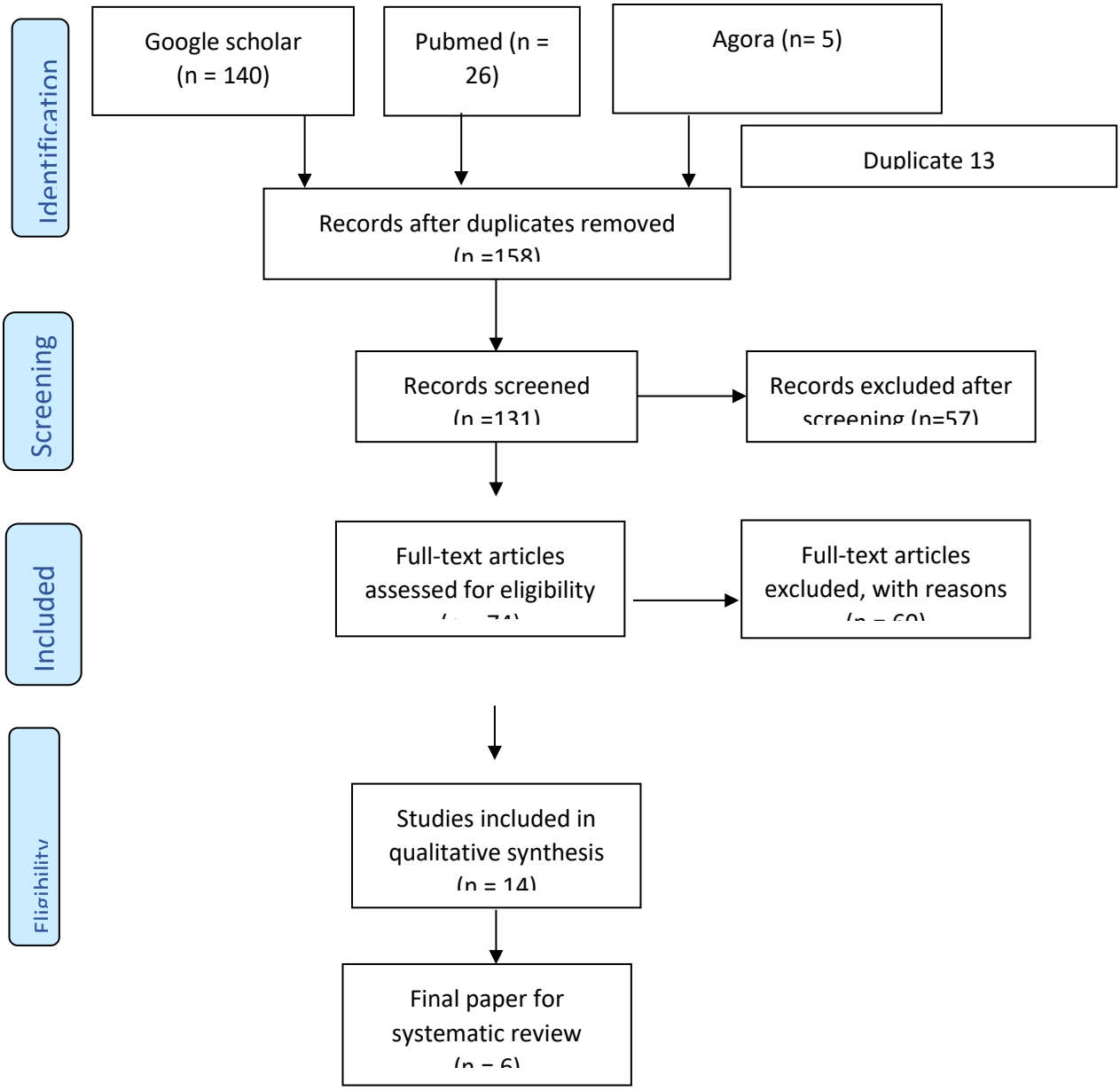

Figure 1: Diagram of included and excluded records of systematic review.

Table1: Characteristics of included studies in the review

Country	Authors	Year of study	Hosts	Tests	Sample (tissue/lait ou autre)	Size of host sample examined	Prevalence of Mycobacterium bovis in animals (%)
Mozambic	Tanner et al 2015	not mentionned	African buffalo (*Syncerus caffer*)	ELISA	Serum	62	5/62 (8.06%)
Mozambic	Tanner et al 2015	Not mentionned	Cattle (Bos indicus)	ELISA	Serum	2445	24/2445 (0.98%)
Zambia	Hang'ombe et al 2012	2006-2010	Cattle (*Bos indicus*)	Culture, PCR	Tissue	315	37/315(0.11%)
Zambia	Hang'ombe et al 2012	2006-2010	Antelope (*Kobus lechwe Kafuensis*)	Culture, PCR	Tissue	15	15/75(0.2%)
South Africa	Sichewo et al 2019	2012-2017	Cattle (*Bos indicus*)	Culture	nasal swab	99	9/99(9%)
Tanzania	Katale et al 2014	2010-2013	Cattle (*Bos indicus* African buffalo (Syncerus caffer), African civet (Civettictis civetta)	Culture, PCR multiplex	Tissue	6	6/606(0.009%)
Zimbabwe	Rosen et al 2017	2014-2016	African elephant (Loxodonta Africana)	MTB Complex antibodies using the ElephantTB Stat-Pak and the DPP VetTB Assay for elephants	whole blood	6	6/35(17.10%)
Tanzania	Katale et al 2017	Not mentionned	African buffalo (*Syncerus caffer*)	Gamma interferon	whole blood	102	2/102(0.01%)

3.2- Prevalence rates

Different species of wild animals are diagnosed for bovine tuberculosis mostly the African buffalo (*Syncerus caffer*) and the percentage of positive bTB reactors in buffalo was 8.06 % using BovidTB Stat-Pak® at the Limpopo National Park Mozambique. In South Africa, forty-one (41) isolates from buffalo, four (4) isolates from lion, two (2) isolates from baboons and one (1) from a warthog were confirmed as *M. bovis*. In Uganda the prevalence of the disease may vary from 5 to 30% in warthogs (*Phacohoeurs africanus*) at the Queen Elizabeth National Park. *M. bovis* was isolated from 11.1% (2/18) migratory wildebeest (*Connochaetes taurinus*) and 11.1% (1/9) topi (*Damaliscus lunatus*) sampled systematically in 2000. Spoligotyping results revealed that one *M. bovis* isolates from one African civet (*Civettictis civetta)* belonged to SB0133 spoligotype. Thirty-five working African elephants (*Loxodonta Africana*) were screened for *Mycobacterium tuberculosis* complex antibodies using the ElephantTB Stat-Pak and the DPP VetTB Assay for elephants. Six of thirty five elephants (17.1%) were seropositive.

3.3- Host Epidemiological Status, Identification of Members of the *Mycobacterium Tuberculosis* Complex, Methods of Diagnosis of *M. bovis*

African buffaloes (*Syncerus caffer*) are regarded as the likely maintenance hosts of bTB usually in Africa; and epidemiological evidence has been gathered to support this view based on the characterization of populations maintaining high bTB prevalence rates. Despite long-term lack of contacts with cattle. Other reported hosts are considered spillover, such as lions, african civet, and elephant. Bovine tuberculosis has been confirmed by culture in 29 free-ranging african wildlife species. Only african buffaloes (*Syncerus caffer*), greater kudus (*Tragelaphus strepsiceros*), and Kafue lechwe (*Kobus leche kafuensis*) are established maintenance hosts (Michel et al. 2009, 2015; Renwick et al. 2007; Munyeme et al. 2010; Clifford et al. 2013), but there are strong indications that warthogs (*Phacochoerus africanus*) and lions (*Panthera leo*) also have the potential to become maintenance hosts (Michel et al. 2015).

The methods of diagnosis of *M.bovis* from the wild life species are not different from those used on cattle; such as culture -the gold standard method, followed by PCR, same for human, cattle and wild life. Followed by interferon gamma assay, rapid lateral flow assay, intradermal tuberculin test, serological assay, inspection, and gps. Certain diagnostics are coupled for more precision such as, culture and PCR; interferon gamma assay, couple to spoligotyping; lateral flow assay, couple to culture, lateral flow assay, couple to interferon gamma assay. GeneXpert, spoligotyping permits to show that there are similarities between *M.bovis*, from wildlife (african buffalo, antelope) and *M.bovis* from cattle or humans. Some are more specific to the species.

3.4- Risk Factors

Most studies conducted in wildlife in Africa, are in Mozambique, Ethiopia, Zambia, Tanzania, Botswana, South Africa, Zimbabwe. Mammals are the wildlife species mostly diagnosed for tuberculosis, such as african buffalo, kafue antelope, whom are considered as maintenance host, potentially responsible for the transmission of *M.bovis* to cattle. Other species are also diagnose such as elephant, warthog, and lion. Information concerning age, sex of animals are not reported. Most of the wild tested animals are adults. One of the factor mentioned concerning the possible transmission of bTB between wildlife and cattle is the sharing of same grazing lands; and concerning cattle and human it is the consumption of unpasteurised milk, aerosol.

Discussion

There are fewer studies done in other parts of Africa as far as wildlife is concerned. It might be because of the no accessibility to wild, specific lack of required equipment, lack of policies on wildlife conservation or expensive requirements of wild life animals. The regional distribution of tuberculosis in Africa's wildlife is mostly in East Africa and Southern Africa. Meanwhile researches on *M.bovis* in cattle and human are more much more available, because bTB is endemic in cattle. Moreover, the disease is of economic, sanitary, and public health importance. Most epidemiological studies done in the wildlife are cross sectional studies, mostly conducted on the field, requiring at some point specific methods of diagnosis, if not for the molecular and gross lesions detection which are main diagnosis done. Approximatively all the included studies were published during the last decade of our review as from 2010 to 2020. A comparatively large number of studies address molecular epidemiology. Overall prevalence rates reported for bovine tuberculosis in *Syncerus caffer* (buffalo) is among the highest recorded for these species worldwide (Katale et al, 2014; Tanner et al, 2015; Katale et al, 2017; Sichewo et al, 2020). Prevalence rates have also been noted in lion, baboon, warthog (Sichewo et al, 2020) Kafue lechwe antelope (*Kobus leche Kafuensis*) (Bengis RG et al, 2004; Munyeme M,et al, 2010), migratory wildebeest (*Connochaetes taurinus*) and 11.1% (1/9) topi (*Damaliscus lunatus*) (S. Cleveland et al, 2005). Among the wildlife species mostly studied we also have the kafue lechwe antelope (Bengis RG et al, 2004; Munyeme M et al, 2010; Hang'Ombre et al, 2012). Most studies did not report about age and sex differences. African buffaloes (*Syncerus caffer*) are regarded as true highly likely maintenance hosts of bTB usually in Africa. Other studies present greater kudus (*Tragelaphus strepsiceros*) as possible likely maintenance host too (De Lisle et al. 2001). The same kafue lechwe antelope considered as high maintenance host posing a potentially high risk of transmission of *M. bovis* to livestock, other wildlife, and to members of the local communities (Munyeme et al. 2010a, b; Malama et al. 2013). Other studies describe the kafue lechwe antelope (*Kobus leche Kafuensis*) as feral reservoirs of bTB in Zambia (Bengis RG et al, 2004; Munyeme M et al, 2010). All other reported hosts are considered as dead end spillover hosts. *M. bovis* were found, along other pathogens such as *Mycobacterium intracellulare* who was the most frequently isolated species (20 isolates, 36.4%) from human, cattle and wildlife followed by *M. lentiflavum* (11 isolates, 20%), *M. fortuitum* (4 isolates, 7.3%) and *M. chelonae-abscessus* group (3 isolates, 5.5%) (Katale et al, 2014) in Tanzania. In South Africa, due to sudden death in meerkats (*M suricattae Suricata suricatta*), inspection was carried out after euthanasia, to discover the cause of death which was *Mycobacterium Suricattae*, (Patterson et al, 2017). They methods used to

diagnose mycobacteria in cattle, and human, are the same ones used for wildlife. Nevertheless at some point of time specific diagnosis like the Elephant TB Stat-Pak and the DPP VetTB Assay for elephants (Rosen et al., 2017), are adopted. Risk factors that govern bTB epidemiology and transmission in wildlife and wildlife–livestock remain largely unknown in sub-Saharan Africa.

Conclusion

The present study, updated information on bovine tuberculosis in Africa's wildlife. This improved the knowledge on tuberculosis. Even though most studies in wildlife are recent, it is noted that, studies are mostly carried out in the east and southern part of Africa. Tuberculosis is present in african wildlife, through non- tuberculous mycobacterial, and zoonotic mycobacteria. These causative agents have already been diagnosed through culture, serological assay, and molecular characterisation. The study reported tangible information on the status of African wildlife as regard tuberculosis. In Cameroun, studies on bovine tuberculosis in wildlife remain unknown. This review is intended to update awareness, and to foster more research on risks transmission of tuberculosis from wildlife to domestic animals, and human.

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Data Avaibility Statement.

The documents we consulted for this systematic review where selected through search engines PubMed, Google Scholar, and Agora. The search criteria were specified in advance and the search string used was the following: ‘‘endemics’’ OR ‘‘epidemiology’’ OR ‘‘risk factors’’ OR ‘‘population at risk’’ OR ‘‘health correlates’’ AND ‘‘bovine tuberculosis’’ OR ‘‘mycobacterium bovis infection’’ AND ‘‘cattle’’ AND ‘‘wildlife’’ AND ‘‘Africa’’.

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Subcellular Localization of β-catenin in Submandibular Salivary Glands in Diabetic Male Rats

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Abstract
Background: Submandibular salivary gland (SMG) is a major salivary gland that is specialized for synthesis and secretion of saliva. The structure of the acini and duct system is impaired in SMG of diabetic subjects. Wnt/β-catenin signaling pathway is one of the regulators of the SMG development and regeneration**. Objective:** Is to assess the distribution patterns of β-catenin in adult male SMG of diabetic rats. **Material and method:** Immunohistochemistry for β-catenin was used to localize β-catenin the glandular tissue. **Results:** β-catenin immunostaining showed different patterns of localization in the mucous membrane, cytoplasm, and nuclei of the acini and duct system in control SMG. Interestingly, robust β-catenin reaction was detectable in the striated ducts masking the nuclei. The excretory duct showed strong cytoplasmic immunostaining and moderate staining of the nuclei; however, some nuclei were masked. By contrast, the SMG of the diabetic rats displayed apparent decrease in the membranous, cytoplasmic, and nuclear β-catenin immunostaining in the granular convoluted tubules, striated and excretory ducts. **Conclusion:** β-catenin immunoreaction is decreased in the duct system of SMG of diabetic rats suggesting impaired role of Wnt/β-catenin in the ducts. Wnt/β-catenin pathway may be target in drug therapy of the SMG in diabetic subjects.
Keywords: submandibular salivary gland, diabetes, β-catenin, rats

1. Introduction

Salivary glands are specialized for synthesis and secretion of saliva. They are classified according to size into major and minor salivary glands. Submandibular salivary gland (SMG) is the second largest gland and secrets about 70 % of total daily amount of saliva in the non-stimulated conditions (Kessler and Bhatt 2018). The SMG secrets mixed type of secretion that is mainly serous in nature. In rats, the SMG consists of seromucous acini which empty the salivary secretion into small intercalated ducts that are hard to be distinguished histologically. These ducts lead to larger ducts lined by columnar cells with basal striation known as striated ducts (SDs). In contrast to humans, rats possess another type of ducts, known as granular convoluted tubules (GCTs) that connects between the intercalated and SDs. The GCTs are more developed in male rats than females (Rins de David et al., 1990). The salivary secretion flows from the GCT to excretory duct (ED) that is lined by pseudostratified columnar epithelium and contains tuft cells with microvilli (Amano et al., 2012).

Diabetes mellitus (DM) is a common chronic metabolic disease. It represents a worldwide threat to human well being and the number of affected patients is expected to be doubled by 2030 (Agarwal and Gupta 2016). Hyposalivation is a complications in the diabetic subjects that leads to swallowing and speech disorders, altered taste, exacerbation in dental caries and increase the incidence of oral infection (Stewart et al. 2016). There is no current effective treatment of hyposalivation in diabetic patients. Since the

molecular changes in diabetic salivary gland is still largely unknown, unravelling these changes is essential for developing new therapeutic approaches for hyposalivation.

The canonical Wnt signaling pathway also known as Wnt (wingless/int)/β-catenin is highly conserved. It is crucial for various cellular processes including cell proliferation, differentiation, polarity and migration (Gajos-Michniewicz and Czyz 2020). The canonical Wnt transduction cascade is essential for stem cell maintenance, and tissue regeneration and renewal (Chidiac and Angers, 2023; Hu and Monga , 2021; Schunk et al., 2021; Takahashi and Shiraishi, 2020; Goessling et al., 2009; Zhang et al., 2008). In absence of Wnt ligand, a complex of protein forms that phosphorylate β-catenin and initiate its degradation. In the active pathway, the Wnt ligand complexes with frizzled receptors and coreceptors lipoprotein receptor related protein 5 and 6. This binding inactivates the destruction complex and β-catenin is accumulated in the cytoplasm followed by its translocation to the nucleus to form active transcriptional complex and activate the target genes (Rim et al., 2022). Members of Wnt signaling pathway and its regulators are expressed in adult salivary glands (Queimado et al., 2008; Sun et al., 2008; Hoffman et al., 2002;). It has been shown that Wnt signaling is active in postnatal mice in the ductal cells of SMG and upregulated during the gland regeneration (Hai et al., 2010). The first step to understand the role β-catenin in SMG of diabetic subjects is to study the distribution patterns of β-catenin in the glandular tissue, thus the study was designed to investigate this expression patterns.

2. Materials and Methods

2.1. Study design and animals

The research ethics committee of Faculty of Dentistry at Minia University gave the study approval (Committee No. 97, decision no. 761) and provides the ethical guidelines of the experimental work. The sample size was determined on the same basis as a previously published study (El Shahawy and El Deeb, 2022). Fourteen adult male, Sprague-Dawley rats (Institute of Graduate Studies and Research, Alexandria University, Egypt, weight 150-200 g) were used in the current study according to the institutional guidelines of experimental animal care and use and ARRIVE guideline (Percie du Sert et al., 2020). The rats were kept in regulated regimes of 12 h light and dark and temperature between 22-23 °C. The rats get water *ad libitum* and fed with standard chow. The animals were classified into two groups: the control (n=7) and the diabetic (n=7) groups.

2.2. Induction of DM

To induce DM in rats of the diabetic group, the animals were fasted overnight followed by administration of single dose (140 mg/kg) of intraperitoneal injection of freshly prepared alloxan (SDFCL, Mumbai, India) dissolved in 4% saline. The controls were injected with saline only. After 3 days, blood sugar level was detected. Rats were classified as diabetic when their levels were over 300 mg/dL and were used in the study (Taiema et al., 2019).

2.3. Sample Preparation and Histology

The SMG specimens were dissected and fixed as previously described in (El Shahawy and El Deeb, 2022). Briefly, the SMGs were fixed in 10% formalin overnight then dehydrated in 70%, 95% and 99.5% concentrations of ethanol, xylene-cleared and followed by paraffin embedding. The specimens were sectioned into thin sections of 5-μm thickness, deparaffinized in xylene and rehydrated in 99.5%, 95% and 70% concentrations of ethanol. Hematoxylin and eosin (H&E) were used to stain the deparaffinized sections for general examination (Life Chemicals group, Alexandria, Egypt**).**

2.4. Immunohistochemistry

Paraffin sections of the SMG were processed for β-catenin immunostaining by streptavidin-biotin method. Briefly, the deparaffinized tissue sections were microwaved in citric acid at pH 6.0 to uncover the antigenic sites. Afterwards, the endogenous peroxidases were blocked by immersing the sections in 3% H202 and non-specific bindings were blocked by bovine serum albumin. Then, the tissue sections were incubated

with β-catenin antibody at 4°C. On the following day, the tissue sections were rinsed from the primary antibody and were exposed to the biotinylated secondary antibody followed by another wash and incubation with streptavidin. The specific reaction sites were visualized after diaminobenzidine incubation as brown precipitate at the specific reaction sites (Reibring et al., 2014). Finally, counterstaining was performed using hematoxylin. Negative controls were done; however the primary antibody was excluded.

2.5. Semi-quantitative Analysis

Semi-quantitative analysis was performed under the light microscope, and histopathological examination of SMG tissue sections was scored. At 40x objective, the SMG parenchyma including the duct system and the acini were assessed. β-catenin staining density of the acini and duct system was scored and expressed as negative (-), weak (+), moderate (++), strong (+++) and robust immunostaining (++++) (Reibring et al., 2014).

2.6. Statistical Analysis

Student t-test was used to analyze the data and to compare between control and diabetic groups. The results were presented in form mean ± standard deviation and difference was estimated as significant at p-value below 0.05.

3. Results

3.1. Alteration of Submandibular Salivary Glandular Tissue in Diabetic Male Rats

Assessment of histological sections of adult rat submandibular salivary gland controls revealed normal histological features. The seromucous acini are spherical with circular nuclei. The GCT are kidney-shaped, lined with columnar cells and possess round basally situated nuclei. The SDs are lined by columnar cells with centrally placed nuclei and prominent basal striation. The ED is located between the lobes of the gland, surrounded by a thin layer of connective tissue and possess wide lumen (Fig. 1).

Consistent with the previous studies (El Shahawy and El Deeb, 2022; Salem et al., 2021; Elias, 2020; Yasser and Shon, 2020; Mahmoud and Mahmoud 2017), the diabetic group (Fig. 1) display seromucous acini with degenerative features; the nuclei were pyknotic and cytoplasmic vacuoles were depicted. The SDs depicted loss of their basal striations and showed vacuolization in their cells. The GCTs exhibited vacuolization and nuclear degeneration. The ED showed nuclear degeneration, vacuolization, apparent thickening of the surrounding connective tissue and stagnation of the mucous secretion (Fig.2; Table 1).

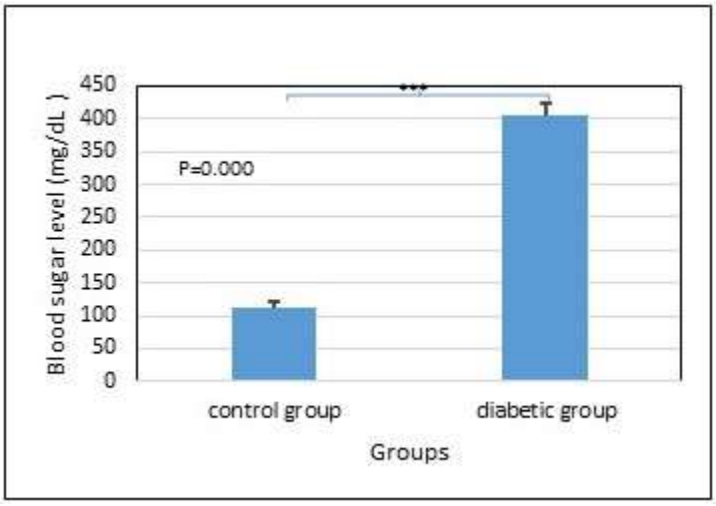

Figure 1. Abnormally high blood sugar level in diabetic rats. Blood sugar level is higher than 300 mg/dL in diabetic group but not in the control group, mean and SD are presented and analyzed by student t-test, n=7 per group.

3.2. Immunolocalization of B catenin in adult diabetic submandibular salivary gland tissue

The SMG of adult male rats displays strong β-catenin immunostaining in the mucous membrane of the acini while the nuclei depict negative reaction. The acinar cytoplasm displays uneven immunostaining pattern. The GCT display strong immunostaining in the membranes, moderate cytoplasmic and nuclear immunoreaction. The SDs show robust cytoplasmic immunoreaction masking the nuclei. The ED depicts strong cytoplasmic and membranous reaction masking some nuclei, however other nuclei show moderate nuclear staining (Fig. 2; Table 1).

By contrast to the control group, the SMG of diabetic rats revealed apparently weak parenchymal β-catenin immunostaining. The seromucous acini show weak immunostaining in the acinar walls, negative nuclear and virtual absence of the immunoreaction in the cytoplasm. The GCT, SDs and ED display weak cytoplasmic, membranous and nuclear immunoreaction (Fig. 3; Table 1).

These data show that β-catenin signaling is altered in the duct system of diabetic rats and suggest that Wnt/β-catenin signaling may be target in treating degenerative changes of diabetic SMGs.

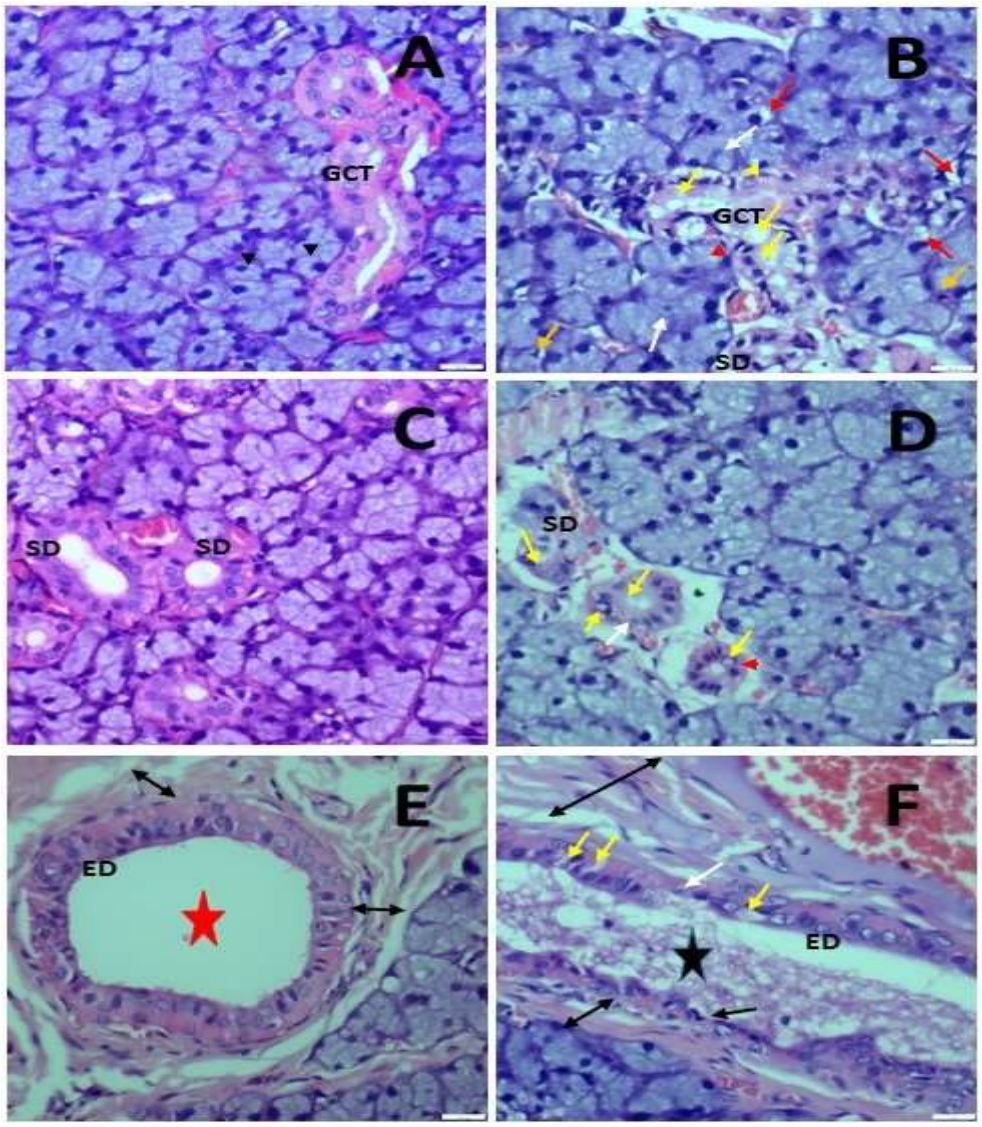

Figure 2. Abnormal histology of adult submandibular salivary gland (SMG) of diabetic male rats. **A, C, E** representative H&E staining images of SMGs of control and diabetic rats (**B,D,F**). (A) The SMG of controls displays seromucous acini with spherical basal nuclei (black arrowhead) and the granular convoluted tubules are lined by columnar cells with rounded basal nuclei (GCT). (C) The striated ducts exhibit basal striations (SD). (E) The excretory ducts lined by pseudostratified epithelium (ED) and surrounded by thin layer of connective tissue (black double arrowhead) and possess wide empty lumen (red star). (B) The seromucous acini of diabetic SMG, display degenerating nuclei (white arrow), vacuolization (red arrow), pyknotic nuclei (orange arrow), granular convoluted tubules (GCT) with vacuolization (yellow arrow), pyknotic nuclei (red arrowhead) and degenerating nuclei (yellow arrowhead). (D) Striated ducts (SD) with vacuolization (yellow arrow), degenerating nuclei (white arrow) and pyknotic nuclei (red arrowhead). (F) The excretory ducts (ED) display cytoplasmic vacuolization (yellow arrow), degenerating nuclei (white arrow), areas of loss of pseudo-stratification (black arrow), lumen with stagnated secretion (black star), and apparent thickening of the surrounding collagen bundles (black double arrowhead). Scale bar: 10 um

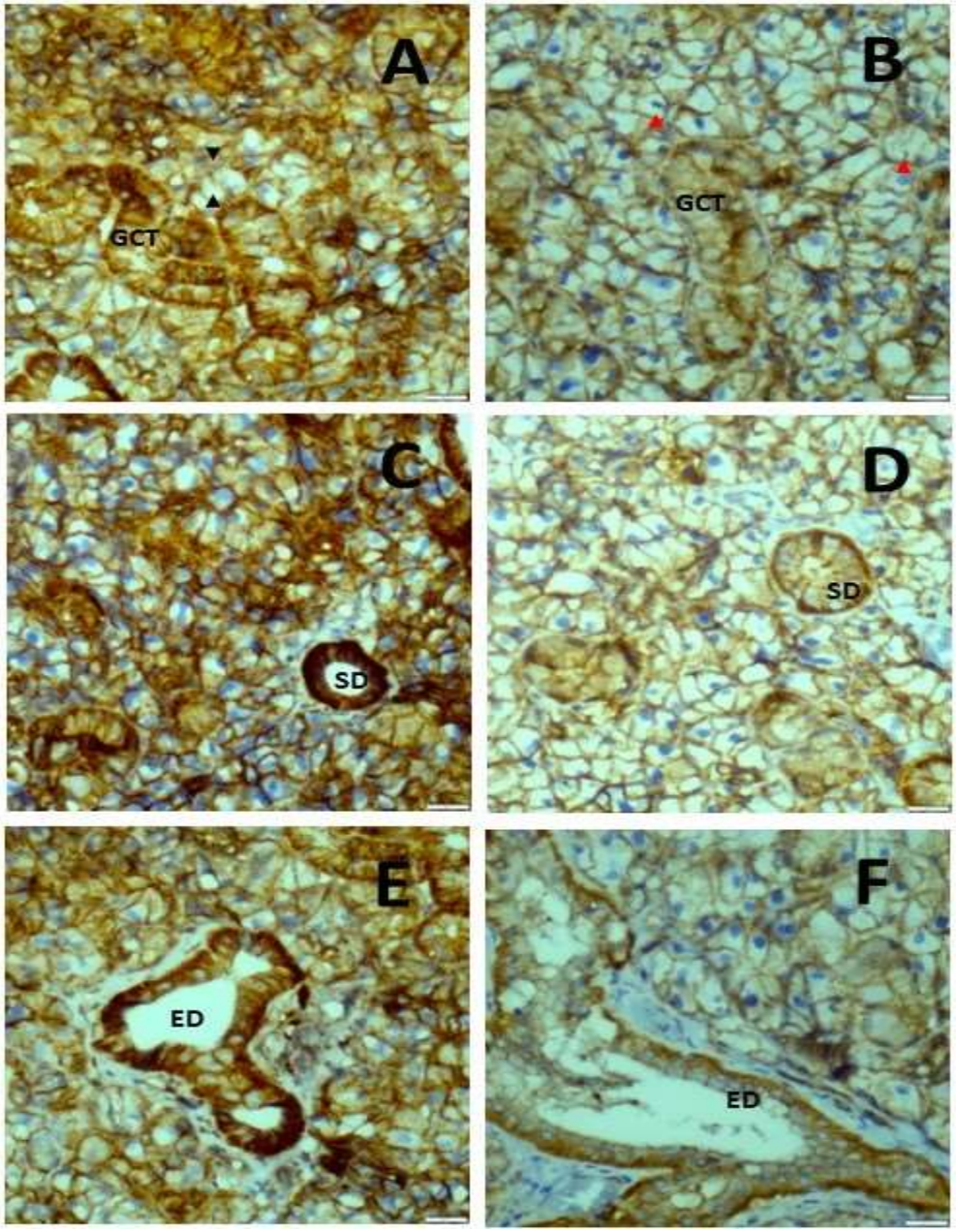

Figure 3. Decreased β-catenin immunostaining in adult submandibular salivary glands (SMGs) in diabetic male rats. Immunohistochemistry of β-catenin showing expression of β-catenin in SMGs of control (A, C, E) and diabetic (B,D,F) animals . Brown color indicates the immunostaining

sites. (A) The seromucous acini of the controls depict strong β-catenin staining decorating the walls of the acini while the nuclei display negative staining and the cytoplasm shows uneven immunoreaction (black arrowhead), the granular convoluted tubules display moderate immunostaining in the cytoplasm and nucleus and strong in the membrane (GCT). (C) Robust immunoreaction is detectable in the striated ducts masking the nuclei (SD). (E) The excretory duct displays strong cytoplasmic and membranous immunoreaction while some nuclei were masked while others depict moderate staining (ED). The immunostaining of β-catenin is apparently weaker in the diabetic group: (B) in the seromucous acini (red arrowhead) and granular convoluted tubules (GCT), (D) striated ducts (SD) and excretory ducts (ED). Scale bar: 10 um (A-F)

Table1: β-Catenin protein distribution in adult male submandibular gland in diabetic rats

Glandular tissue	cytoplasmic	Membranous	Nuclear
Seromucous acini			
Control acini	(A)	+++	-
Diabetic acini	-	+	-
Granular convoluted tubule duct			
Control duct	++	+++	++
Diabetic duct	+	+	+
Striated duct			
Control duct	++++	++++	(B)
Diabetic duct	+	+	+
Excretory ducts			
Control duct	+++	+++	(C)
Diabetic duct	+	+	+

Negative (-), weak (+), moderate (++), strong (+++) and robust (++++) immunostaining. (A) The immunostaining in the cytoplasm of seromucous acini is likely non-specific as the pattern is uneven. (B) Robust β-catenin immunostaining masking the nuclei of striated ducts. (C) Strong β-catenin immunoreaction masking some nuclei of the ED while others depict moderate nuclear staining

4. Discussion

Male rats´ submandibular salivary glands (SMGs) are characterized by larger and numerous GCT compared to females (Rins de David et al., 1990). Male rats injected with alloxan developed diabetes as indicated by the analysis of blood glucose, since alloxan leads to destruction of β-cells of the pancreas (Karami et al., 2022).

The SMG of diabetic rats showed degenerative changes and vacuolization of the acini and duct system, which confirm previous studies (El Shahawy and El Deeb, 2022; Salem et al., 2021; Elias, 2020; Yasser and Shon, 2020; Mahmoud and Mahmoud 2017). These alterations could be due to hyperglycemia in diabetic rats which aggravates the oxidative stress (Mednieks et al., 2009; Anderson et al., 1994). The antioxidant capacity is reduced in diabetes due to changes in the antioxidant enzyme activities and excessive reactive oxygen species (ROS) formation (Bonnefont-Rousselot et al., 2000). Enhanced ROS disrupts the cellular metabolism and destroys the macromolecules (Lushchak, 2014). The ROS damages all biomolecules including RNA, DNA, lipids, carbohydrates, and proteins leading to impairment of biological cell functions (Knaś et al., 2016). Furthermore, the free radicals released due to hyperglycemia lead to abnormalities in the phospholipid bilayer of the cell membrane changing its properties and causing membrane rupture (Potphode et al., 2018). It has been reported that cytoplasmic vacuolization may be due to accumulation of lipids that were dissolved during the processing of the specimens (Yasser and Shon, 2020).

β-catenin is one of the components of the adherence junctions in simple epithelia. It is crucial for the connection of E-cadherin to the cytoplasm which plays role in differentiation of the acinar cells (Patel and Hoffman, 2014; Peifer et al., 1994). In this study, β-catenin immunostaining was observed to be diminished in the plasma membrane of SMG ducts and acini of diabetic rats suggesting disturbance in the tissue homeostasis.

Higher intensity of β-catenin immunostaining was observed in the duct system of SMG of control rats. This is consistent with previous findings (Hai et al., 2010) that shows active β-catenin signaling in the SMG ductal epithelia.

The results of the present work show that the altered duct system including the GCTs, SDs, ED of SMG of diabetic rats displays lower staining intensity of β-catenin compared to their controls. It has been shown that Wnt/β-catenin inhibition for longer duration impairs the postnatal SMG development especially GCT (Hai et al., 2010).

The SDs play role in conduction and modification of the primary saliva. They reabsorb sodium and chloride release epidermal growth factor (EGF) and kallikrein. In addition, SDs are impermeable to water thus the final secreted saliva is hypotonic in nature (Contreras-Aguilar and Gomez-Garcia 2020). The GCT synthesizes and secretes several biological molecules including nerve growth factor, EGF, kallikreins, renin and proteases, and these molecules are crucial for the repair and protection of the mucous membranes. The tuft cells of the excretory duct could be associated with the sensation, resorption, secretion of the salivary production (Amano et al., 2012). Components of Wnt signaling pathway and regulators are expressed in the salivary glands. Wnt4, Wnt5a and secreted frizzled related protein 2 are expressed in adult SMG (Hoffman et al., 2002, Daa et al., 2004). Inhibition of Wnt signaling in young adult mice interferes with the postnatal SMG development with GCT being most affected. Thus, Hai et al, suggest marginal level of β-catenin signaling is required for postnatal development of GCTs (Hai et al., 2010). In addition, Wnt/β-catenin play role in SMG branching morphogenesis (Häärä et al., 2011). It is known that Wnt signaling pathway is crucial for organ renewal and regeneration. During the processes of regeneration, Wnt signaling increases in the stem cell compartment. Impairing Wnt signaling interferes with the process of regeneration while activating the pathway enhances the regeneration (Goessling et al., 2009; Goessling et al., 2008). Hai et al., showed that Wnt/β-catenin is upregulated during SMG duct regeneration and that it plays role in the molecular control of the process of regeneration. Furthermore, forced activation of Wnt/β-catenin signaling expands the stem cell compartment and induces ductal proliferation indicating that Wnt/β-catenin signaling is sufficient to induce regeneration of SMG (Hai et al., 2010). Taken together, these data suggest disturbance in Wnt/β-catenin which may lead to impaired regenerative capacity in the duct system of diabetic rats.

Limitation of the current study is that it was performed on male rats only, while females were not assessed with possible role of hormonal interaction with β-catenin signaling cascade in diabetic SMG of females. Further investigation of canonical Wnt signaling pathway components´ role and its interaction with other pathways is of importance to understand the molecular regulations in SMG of diabetic subjects.

5. Conclusion

The Wnt/β-catenin signaling regulates many cellular processes and is crucial for tissue regeneration and renewal including SMG (Hai et al., 2010; Goessling et al., 2009; Goessling et al., 2008). The purpose of the current work was to show the distribution patterns of β-catenin in the SMG tissue by using immunohistochemistry. The study results revealed decreased β-catenin protein in the duct system including GCTs, SDs, and ED, suggesting disturbance in β-catenin signaling in these ducts.

6. Acknowledgements

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Abbreviations

DM	Diabetes mellitus
ED	Excretory duct
EGF	Epidermal growth factor
GCTs	Granular convoluted tubules

H&E	Hematoxylin and eosin
ROS	Reactive oxygen species
SDs	Striated ducts
SMG	Submandibular salivary gland
Wnt/β-catenin	Wnt (wingless/int)/β-catenin

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The Synergistic Effect of Combining Pituitary Gland and Ovaprim Hormone on Latency, Fertility, Hatchability, and Survival of African Clariid Catfish - *Clarias Gariepinus*

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Abstract
The hatchability of *Clarias gariepinus* using pituitary extract and Ovaprim hormones was carried out at the Hatchery unit of the Fisheries Department, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria. One male to two females were used across all batch treatments (T1-T3). Batch T1 was administered 100% Ovaprim, T2 received 100% Carp Pituitary Extract-CPE, T3 a combination 50% ovaprim and 50% CPE. T4 was without any hormonal treatment. The latency periods were 12h, 16h, 10h and 0h at 27.8ºC, 26.9ºC, 27.5ºC and 26.6°C, respectively, for T1, T2, T3 and T4. T3 had the highest fertilization (78%) and hatchability (85%) rates, followed by T1 (72%, 66%), T2 (61%, 56%) and lastly T4 (26%, 28%). The hatching duration was 18h, 20h, 20h, and 23h with T3, T1, T2 and T4, respectively. Survival rates were in the order: T3 (87.77%), T1 (84.44%), T4 (81.11%) and T2 (76.66%). T3 had higher mean weight gain (0.39g) and specific growth rate (10.07%) compared to T2 (0.39g, 9.63%), T4 (0.33g, 9.54%) and T1 (0.32g, 9.44%). Economically, Ovaprim (T1) cost (₦700 or 1.71USD) while CPE (T2) cost ₦1382.4 (3.37USD). The combination (T3) had the best fertility, hatchability, survival rates, and best growth performance of African Clariid Catfish *Clarias gariepinus* fingerling production and is hereby recommended to fish breeders.
Keywords: catfish, breeding, synthetic, pituitary, combined hormone, No hormone

Introduction

The demand for fish is geometrically growing along with population growth, urbanization and increasing wealth globally to the tune of USD 416 billion in 2020 (FAO, 2022a). FAO (2022a) reported that per capita fish consumption will increase fastest in rich countries and in some parts of the world, such as China, where aquaculture thrives, leading to growing regional disparities between supply and demand. As a result, the increase in population and continued depletion of natural resources resulting from exploitation in developing countries, and the exponentially rising cost of living have made the rates of hunger and poverty surge to very high levels (Akinrotimi *et al*., 2015a; Akinrotimi *et al*., 2007; Stanley *et al*., 2003). The cost and demand for food especially the dietary fish protein has also risen considerably. In view of this, agriculture and science have been coming up with better ways of accelerating productivity to meet the population needs (Akinrotimi *et al*., 2015b; Anyanwu *et al*., 2007; Hogsetle, 1999). Most worrying, however, are the declines in projected consumption for sub-Saharan Africa, where levels of food and nutrition insecurity with poverty are alarming.

Fish make up such a high proportion of animal-source foods (32%). It is indicated that total fisheries and aquaculture production besides algae has drastically expanded in the last seven decades moving from 19 million tonnes (live weight equivalent) in 1950 to all-time high of about 170 million tonnes in 2018 with an annual growth rate of 3.3 percent (FAO 2022a). If algae production is added to that of the aquatic

animals, fisheries and aquaculture production will reach 214 million tonnes in 2020, with an overall growth of 0.3 percent (FAO 2022a). Overall, aquaculture had already overtaken capture fisheries as the primary source of aquatic production since 2013 and has reached its share of total production of 57 percent in 2020 (FAO 2022a). The stagnation experienced in the last two years is mainly associated with a slight decline in capture fisheries to 4.5 percent in 2019 in contrast to the peak of 96 million tonnes in 2018 with additional decrease by 2.1 percent in 2020 FAO 2022a). According to FAO (2022b), the long-term trend in global capture fisheries continued to be relatively stable with catches generally fluctuating between 86 million tonnes and 93 million tonnes per year since the late 1980s. Though in 2020, the global capture fisheries production (excluding algae, SOFIA 2022) was 90.6 million tonnes -a fall off 4.0 percent in comparison to the last three years. This decrease was due to the disruption in fishing operations resulting from the COVID-19 pandemic, the fluctuating catches of pelagic species and the ongoing reduction in China's catches (10 percent lower in 2020 as against average of the previous three years) (FAO 2022a, b). Global fish sales from capture fisheries in 2020 was USD 131 billion compared to USD 265 billion from aquaculture indication that capture fisheries will therefore not be able to meet the growing global demand for aquatic food (FAO 2022a).

The high demand for fish fingerlings in the high growing aquaculture industry has stimulated the need for artificial propagation of cultured warm water fisheries (FAO, 2007). African catfish (*Clarias gariepinus*) is a hardy species for aquaculture purpose. It is widely accepted in the tropics and commands good commercial values, even though, the species hardly breeds naturally in captivity. All year-round efforts are to be intensified on research for its artificial seed production (Oguntuase & Adebayo, 2014)). Sex steroids in female fish perform major roles in Oocyte maturation, ovulation, and spawning. Synthesis of Vitellogenin and increase in ovarian size during final Oocyte maturation is controlled by 17b-estradiol, which is directly related to gonad somatic index (Coccia *et al.*, 2010). Although a trend exists to apply alternative synthetic hormone substances to induce spawning in catfish and other cultured fishes, hypophysation still remains the most common technique. *Clarias* pituitary extract (CPE) and luteinizing hormone-releasing hormone analogue (LHRHa) are two well-known hormones for controlling ovulation in Channel catfish (Forbes, 2013). Among the most important advancement in the field of aquaculture to tackle this challenge in recent times, is the development of techniques to induce reproduction in fish. *Clarias gariepinus* shows a seasonal gonadal maturation which is usually associated with the rainy season. The maturation processes of *Clarias gariepinus* are influenced by annual changes in water temperature and photoperiodicity and the final triggering of spawning is caused by a rise in water level due to rainfall (de Graaf *et al*., 1995). The pituitary gland extract has advantages of better rate of fertilization and hatching, better conditions for growth and survival of larvae (Woynárovich and Horváth, 1980). In addition, it enhances free release of eggs from the genital papilla of female broodstock and stimulates the release of hormones into the blood circulatory system which induces ovulation and production of mature eggs (Oyeleye *et al*., 2016). Also, African catfish pituitary hormone is said to be readily available and cheaper than any other hormone (Adebayo and Popoola, 2008) and can be prepared in a suspension (Fagbenro *et al*., 1998). However, the extraction of the pituitary gland from other donor species is a challenge. For instance, Common carp (*Cyprinus carpio*) pituitary gland materials are not easily accessible (Olaniyi & Akinbola. 2013).

The use of synthetic hormones in African catfish particularly in female fish is now popular as a means of artificially inducing the female fish to ovulate. These include synthetic gonadotrophin releasing hormone analogues (GNRH-a) which are administered to the female brood stocks (Oyelese, 2006). Gonadotropin Releasing Hormone analogue (GnRHa) is now the best available biotechnological tool for the induced breeding of fish. These include the following which have been used to induce breeding successfully: Ovaprim, Ovatide, Ovaryprim, Ovopel, Ovupin-L, Ovulin, Dagin and Aquaspawn (Ngueku, 2015; Zhuo, *et al*., 2011; Nwokoye, et al., 2007; Cheah and Lee. 2000; Brzuska & Adamek. 1999). Among GnRHa are

Ovaprim and Ovatide which contain salmon gonadotropin releasing hormone analogue and domperidone (SGnRHa + Domperidone) often used for spawning induction in catfishes to get quality seed (Sahoo *et al*., 2014). In addition, for large scale production in hatcheries, the use of hormones may be the solution. However, some of these hormones particularly Ovaprim is usually expensive (Khan *et al*., 2006, Madu, 2006, Olubiyi *et al*., 2005). Ovaprim is reported to significantly increase ovulation in African catfish (Sharaf *et al*., 2012) and aid spawning in the matured female catfish. According to Watson *et al*. (2009), the average success rates of 50% ovulation, 54% spermiation and 1.3% mortality were recorded after injection of different species with Ovaprim. Also, it has been used successfully for hypophysation in different families of fish like the Cyprinidae (Hill *et al*., 2005), Characidae and Cobitiidae (Yanong, Martinez &Watson, 2009). The objective of the study is to determine the best hormonal approach towards improvement in breeding of African catfish fingerlings. Hence, this research work aimed at improving on the latency period, egg fertilization, hatching rate, and survival of hatchlings of African clariid (*Clarias gariepinus)* uses pituitary and Ovaprim hormone singly, in their combined form or none to breed.

Methods

The Study Area

This research work was conducted in the Department of Fisheries and Aquatic Resources Management of Michael Okpara University of Agriculture, Umudike, Nigeria which lies between latitude $5^0$29'N and $7^0$33'E. The average temperature of the area is 26^0C, with minimum of 22^0C and maximum of 32^oC. It is 122m (499ft) above sea level with an average rainfall of 21698.8mm obtainable within 148-155 days. The relative humidity ranges from 50% to 95%. The town is located within the humid rainforest zone characterized by long duration of rainfall of 7-12 months and short period of dry season.

Brood Stock Selection

A total of twelve adult (brood stocks) African catfish (four males and eight females) was used in this study. The brood stocks of the *Clarias gariepinus* were obtained from a renowned fish farm along Ussa Road, Abia State, Nigeria. The mature gravid females were selected based on well distended swollen soft abdomen, reddish vent, and easy extraction of few eggs by gently depressing the fish abdomen using the finger. Mature males were selected based on their reddish pointed genital papillae (Ajah, 2019; de-Graff & Jonsen, 1996,).

Transportation and Acclimatization of Procured Brood Stock

The brood stocks of *Clarias gariepinus* were acclimatized in concrete holding tanks of the Fisheries Department Farm 24 hours before artificially induced breeding was carried out. Both males and females were acclimatized in separate concrete ponds of 8×8×5 ft.

Sources of Hormone

Non-Synthetic Hormone: Dried Carp Pituitary Extract (CPE) was purchased from a local supplier in Calabar, Nigeria.

Synthetic Hormone: Ovaprim, produced by Syndell laboratories, Canada has been in use as an inducer for fertility since the early 1990s till today. It was purchased at Abayi Road, Aba, Abia State, Nigeria.

Hormone and Pituitary Gland Injection

Single application of ovaprim as described by (Ndimele & Falayi, 2012; Nandeesha, *et al* 1990;) was adopted. Two female brood stocks and one male were injected intramuscularly (above the lateral line, towards the tail (Figure 1) with ovaprim, at the recommended dosage of 0.5ml per kg body weight of female fish and half dosage for male (Table 1) according to Legendre (1986) and was designated as T1. Two other females and one male were injected with the recommended dosage based on their weight with 100% Carp Pituitary Extract-CPE and was tagged T2. T3 batch had a combination of 50% ovaprim and 50% CPE. T4

group which served as control had no hormonal treatment on both matured male and gravid female catfish. Upon completion of all injections at intervals of eight (8) minutes in between batches, the batches were conditioned in separate plastic tanks for a latent period of 12 hours though examined hourly from the 8th hour post injection of hormone to ascertain readiness for stripping.

Figure 1: Shows intramuscular injection of hormone on catfish

All the females were first tranquilized using anaesthetizing agent (MS222) before stripping to avert any wound and thence reduce stress/pain. Thereafter, the stomach of the two gravid and matured females were pressed gently to release the matured eggs while the males were sacrificed to extract the milts.

Table 1: Dosage of Synthetic (Ovaprim) and Non-Synthetic (CPE) Used in Female and Male Broodstock Treatments.

Treatments	ABW(g) Female	Hormones	Dose/Kg Body Weight (ml)	ABW(g) Males	Hormones	Dose/Kg Body Weight(ml)
T1	1450	100% ovaprim	0.4	750	100% ovaprim	0.3
T2	1500	100% pituitary	1.5	650	100% pituitary	0.8
T3	1350	50% ovaprim + 50% pituitary	0.19 + 0.38	550	50% ovaprim + 50% pituitary	0.25 + 0.5
T4	1350	None	-	750	None	-

ABW= Average Body Weight (g)

Milt and Egg Collection

The milts were collected by sacrificing the males. The two lobes of the male testes were removed, cleaned with tissue paper, and kept in a clean dry labelled petri dish. The abdomen of the female was well cleaned with tissue paper to avoid contact between the eggs and the water. Then the females were stripped of their eggs by a gentle application of pressure on the abdomen to release the eggs. The eggs were collected in a clean dry labelled plastic bowl.

Artificial Fertilization, Incubation and Hatching of Eggs

Dry fertilization method was used. The testes of the male were cut using a dissecting blade and the milts were squeezed out. Saline solution (0.9%) was then added to the milts to facilitate fertilization after which the semen was used in fertilizing the already stripped eggs.

Seven hundred (700) eggs were used for fertilization in each treatment. The number of eggs were estimated using gravimetric method (no of eggs/g). The translucent eggs containing embryonic eyes at the time of

polar cap formation (about 20 minutes after fertilization) were considered fertilized and counted to calculate percentage fertilization. Opaque eggs were considered unfertilized.

Incubation and hatching of eggs were carried out in four (4) hatching tanks of 600 liters capacities each filled with 500 liters of water and containing incubation nets of 1mm mesh size to serve as substrate attachment for eggs. The fertilized eggs were evenly spread on the incubation nets in the tanks and water temperatures kept between 26.7- 27.8ºC.

Experimental Design

The experiment was arranged as a complete randomized design (CRD) with four (4) experimental treatments in triplicates. In this study, 8 female brood fish and 4 male brood fish making a total of 12 brood stocks were used for the study. That is, two females to one male for each of the treatments. Brood fish were weighed before and after stripping out the eggs. The weight of the roes and eggs were taken using a sensitive weighing balance. The hatchlings of each of the treatments were allowed for a period of 3 days for endogenous feeding. Then they were fed with decapsulated artemia for 7 days after which, 30 samples were randomly selected from each of the treatments and were transferred to the experimental layout where they were raised for the period of 30 days.

Feeding Regime

The frys were fed at a fixed feeding rate of 3% body weight four times daily between 07:00 and 23:00 hours at regular intervals for a period of 30 days with Coppens Feed.

Water Quality Parameters

Water quality parameters such as pH, temperature, and dissolved oxygen were monitored daily for the first two weeks and later weekly while ammonia was monitored weekly. A smart DO meter, an accument instrument for temperature and pH were used while some water samples were taken and fixed *in situ* for ammonia determination. Each water parameter was taken thrice from each replicate and the average was recorded.

Fish Weight and Length Measurement

The initial body weight and length of each set of fish was measured using a sensitive weighing balance and meter rule, respectively, before stocking. Subsequently, bulk- weighing and length of fish in each treatment were done after every 7 days.

Determination of Reproductive Success Parameters

The following formulae (Lambert, 2008) were used:

Fertility rate:

$$\% \text{ Fertility} = \frac{\text{Number of fertilized egg} \times 100}{\text{Total number of eggs counted.}}$$

Hatchability rate:

$$\%\text{Hatchability} = \frac{\text{Number of eggs hatched (fry)} \times 100}{\text{Number of eggs incubated.}}$$

Survival rate:

$$\%\text{survival} = \frac{(\text{total no of larvae} - \text{no of dead larvae}) \times 100}{\text{total no of larvae}}$$

Analysis of Fish Growth

The growth expressed as mean weight gain, specific growth rate and survival rate were determined as follows:

Mean Weight Gain (Mwg)

The fish mean weight gain was determined as the difference between the final mean weight of the fish at the end of the experiment and the initial mean weight in grams (Duncan, 1955).

MWG = (W-1) -(W-0)/n
Where: W = initial mean weight; W_1= final mean weight; n = number of fish in the tank

Specific Growth Rate (Sgr)
This is the mean percentage increase in body weight per day over a given time interval (Abdulraheem, *et al*., 2012). In this study, the time interval was 30 days.

$$SGR = \frac{InW2 - InW1}{T} \times \frac{100}{1}$$

Where, W2= final weight
W1= initial weight
T= duration of culture period
In = natural Log

Cost Analysis

i. Cost of hormone used in T1 = ₦ 1000 x 0.7ml = ₦ 700
ii. Cost of hormone used in T2 = ₦ 600 x 2.3ml + ₦ 0.6 x 4 = ₦ 1380 + ₦ 2.4 = ₦ 1382.4
iii. Cost of hormones used in T3 = ₦ 1000 x 0.44ml + ₦ 600 x 0.88ml + ₦ 0.6 x 1.5ml = ₦ 440 + ₦ 528 + ₦ 0.9 = ₦ 968.9
iv. Cost hormone used in T4= Nil

Statistical Analysis
The results obtained were subjected to one way analysis of variance (ANOVA). The statistical differences between the treatment means were separated using Tukey's test (SPSS, 2010).

Results
Water Quality Parameters Taken from the Experimental Tanks for the Hatching Period.
The temperature ranged from 26.6 – 27.8ºC, while pH was from 6.21 – 7.23, and Dissolved Oxygen between 5.2 and 5.7mgO_2/l (Table 2).

Table 2: Water Quality Parameters Taken from the Experimental Tanks during Hatching Period.

Parameters	T1	T2	T3	T4
Temperature(°C)	27.8	26.9	27.5	26.6
pH	7.10	7.23	6.52	6.21
Dissolved Oxygen (mg/l)	5.3	5.2	5.7	5.6

Induced Ovulation and Spawning of *Clarias Gariepinus* Using Synthetic (Ovaprim) and Non-Synthetic (CPE) Hormones are illustrated in Table 3. Average Body Weight (ABW), mean egg weight, fertility rate, hatchability, survival, hatching/incubation periods, and latency periods of all the treatments are presented in Table 3. T3 recorded the highest survival rate while T2 was lowest. The highest hatchability was recorded with T3 followed by T1, and T2 while the lowest hatchability was recorded in T4. The highest fertility rate was also recorded in T3 and the lowest was in T4. Latency period which is the period between the time of injection and striping was also put into consideration. The latency periods of the fish in T1, T2, T3 and T4 were 12, 16, 10hours and O hours (the control) respectively. Those in the combined effects T3 was shortened because of the synergy of the two hormones while T1 remained normal. Thus, T2 had the longest latency period of 16:00 hours while the shortest was recorded in T3. Also, the hatching periods of the treatments varied ranging from 18.00hrs-23.00hrs. The average body weight of gravid female ranged from 1350-1500g.

Table 3: Induced ovulation and spawning of *Clarias gariepinus* using synthetic (ovaprim) and non-synthetic (CPE) hormones.

Parameter	**T1**	**T2**	**T3**	**T4**
ABW(g)	1450	1500	1350	1350
Fertility Rate (%)	72.00	61.00	78.00	26.00
Hatchability (%)	66.00	56.00	85.00	28.00
Hatching/Incubation Period(hrs)	20.00	23.00	18.00	20.00
Latency(hrs)	12.00	16.00	10.00	-
Survival (%)	84.44	76.66	87.77	81.11

Percentage Survival Rate

The percentage weekly survivors per replicate per month are shown in table 4.

Table 4: Weekly survival of fries

Treatments	T1			T2			T3			T4		
Replicates	R1(%)	R2(%)	R3(%)	R1(%)	R2(%)	R3(%)	R1(%)	R2(%)	R3(%)	R1(%)	R2(%)	R3(%)
Week1 Sr	93.3	100	100	100	93.3	93.3	100	100	100	100	96.7	100
Week2 Sr	96.7	100	96.7	96.7	93.3	93.3	97	96.7	100	100	96.7	96.7
Week3 Sr	96.3	97.3	96.7	90	93.3	96.3	96.7	93.7	86.7	96.7	96.7	90
Week4 Sr	96.3	90	90	90	93.3	96.7	933	90	96.7	93.3	96.7	90
1 Month	83.3	86.6	83.8	76.6	73.3	80	86.6	83.3	93.3	80	83.3	80

Key: SR- Survival Rate

The Rate of Growth of Fry at Seven Days Intervals for 30 Days.

Table 5 shows the growth of fry taken after every seven days for 30 days, the growth parameter taken for the fry included the initial and final weights, the mean weight gain, and the specific growth rate. The initial weight was the same (0.020g) for the hatched larva. The final weight gained recorded were 0.34g recorded in T1, 0.36g in T2, 0.41g in T3, and 0.35g in T4. The fry in T3 had the highest final weight of 0.41g while T1 was the lowest with 0.32g at day 30. The mean gain weight of T3 was obviously higher with 0.39g and lowest with T1 with 0.32g induced with 50% ovaprim and 50%CPE and 100% ovaprim respectively. The specific growth rate was also highest (10.07) in T3 and least (9.44) in T1.

Table 5: Growth parameters of the fry.

Parameter	**T1**	**T2**	**T3**	**T4**
Initial weight(g)	0.020	0.020	0.020	0.020
Final weight(g)	0.34	0.36	0.41	0.35
Mean weight gain	0.32	0.34	0.39	0.33
Specific growth rate	9.44	9.63	10.07	9.54

Cost Analysis for Hatchability of *Clarias gariepinus* Using Ovaprim and CPE

Table 6: Reveals the average cost of the hormones used per treatment in this study. Treatment 4 recorded zero cost of production followed by T1 while Treatment 2 recorded the highest cost of production.

Table 6: Average cost analysis of hormones used per treatment.

Parameters	T1(₦)	T2(₦)	T3(₦)	T4(₦)
Ovaprim	700	0	440	0
Pituitary	-	1380	528	0
Normal Saline	-	2.4	0.9	0
Total	700	1382.4	968.9	0

Fish Length-Weight Relationship

T1, T2, T3, T4 had lengths of 1.41±0.78, 1.28±0.76, 1.74±1.05, and 1.37±0.87 cm, and weights of 0.14±0.16, 0.13±0.15, 0.19±0.23, and 0.13±0.15 g, respectively (table 7). Consequently, a negative allometry which indicates that the increase in the length at this stage of growth was not proportional to weight gain as observed. This is attributed to their stage of growth and feed utilization by the fries. Conversely, the coefficient of determination(r) which helps to determine the rate of growth, was positive indicating that the fry growth rate is normal.

Table 7: Length weight relationship of the experimental fish.

Treatments	Length(cm)	Weight(g)	α	B	Allometry	R	Inference
T1	1.41±0.78	0.14±0.16	0.9946	0.7881	0.7881	1	+
T2	1.28±0.76	0.13±0.15	0.996	0.8258	0.8258	1	+
T3	1.74±1.05	0.19±0.23	0.9976	0.7885	0.7885	1	+
T4	1.37±0.87	0.13±0.15	0.9986	0.8266	0.8266	1	+

Discussion

The impact of both synthetic (ovaprim) and non-synthetic (Carp pituitary extract, (CPE) hormones singly or in combination as well as non-hormonal treatment on the latency period, fertility, hatchability, growth and survival of the hatchlings and cost-effectiveness on *Clarias gariepinus* was established. The results showed the importance of inducement on breeding efficiency using synthetic and natural hormones on the targeted species to aquaculture sustainability. Average Body Weight of gravid females and males used in each treatment respectively ranged from 1350g-1500g and 550g-750g. Spawning occurred within 12h, 16h and 10h after injection of the hormones at temperatures of 27.8ºC, 26.9ºC and 26.6°C, respectively for ovaprim, pituitary and their combinations which compare favourably with Gomina (2011) who had a latency period of 10h using varying levels (0.1, 0.3 & 0.5ml/kg) of ovaprim. While the latency period, fertility rate, hatchability, incubation period, percentage survival and SGR, between Ovaprim and pituitary varied considerably, Onyia *et al*. (2021) opined that there is no difference in the use of ovaprim and pituitary extract, and that each can supplement for the other reason being that both contain Gonadotropin releasing hormone analogue (GnRTHa) hormone and Domperidone (SGnRHa _+Dompeidone) (Sahoo, *et al*., 2014) though not in the same proportion..

All treatments were significantly ($p<0.05$) different from each hormonal application. The highest temperatures of 27.8°C which occurred in T1(Ovaprim) at 0.4 ml/kg in ovaprim hormone did not yield the highest fertilization and hatchability rates rather the T3 (50% ovaprim + 50% CPE hormone) had the highest fertilization and hatchability rates of 78%, 85% respectively at temperature of 27.5ºC. These results agree with the previous studies of Oyelese (2006) who observed that at higher temperatures, fertilization and hatchability gave the best results. The findings are equally in agreement with Amaechi and Solomon (2015) who opined that optimum temperature ranges for fertilization and hatchability was between 26-27 °C. Gomina (2011) also reported that the best temperature for fertilization and hatchability was 25.9°C while Alemayehu (2015) had 25.10-26.12^{0}C. Akombo *et al.* (2018) had temperature ranges of 20.66ºC to 23.0ºC and 20.83ºC to 22.83 ºC, respectively, using Ovaprim and Ovatide for hatching using same species. The lower fertilization and hatching rates observed in T4 (without hormonal injection) was not unconnected

with the difficulty inherent in stripping out enough eggs and the readiness of the eggs fertilization hence the need for hormonal injection. culture temperature (26.6oC) was within the optimum temperature ranges for breeding (Akombo, *et al* 2018; Alemayehu 2015; Gomina 201l)

This study further revealed that ovaprim had a higher hatchability rate (66%) compared with pituitary extract (56%), although the highest hatchability rate (85%) and survival rate (87.77%) was recorded in the combined therapy while the lowest hatchability rate (28.0%) was recorded where no inducement (T4) was carried out. By implication, hormonal application is not just needed but in right proportion and better still, in a combined form to arrive at the best breeding output. Similarly, the survival rate was higher in ovaprim (84.44%) compared with pituitary (76.66%) but lowest for the non-hormonal treated batch. This agreed with Nayak *et al.,* (2001) who reported higher hatching rate of 96% using Ovaprim. It also agreed with the study reported by Belal Hossain *et al.,* (2012) that hatching rate of 76.9% in eggs spawned from Ovaprim induced individual fish compared to 72.7% in ACPE induced fish. This study differs from the finding of Olaniyi and Akinbola (2013) who reported higher survival rate from pituitary gland extract. The variation could be because of seasonal temperature variation between where this research was conducted (Abia State, Nigeria) and where these other research works were conducted or the state of the Ovaprim hormone used.

The result also indicated that there was difference in the cost of using ovaprim compared to pituitary extract which cost ₦700 (1.7005USD) and ₦ 1382.4 (3.587USD) respectively making it more economical to use ovaprim than using pituitary extract. Similarly, Olaniyi and Akinbola (2013) arrived at almost the same value of cost of production with ACPE when compared to ovaprim.

Conclusion

The synergistic effect of half the recommended doses of Ovaprim and pituitary gland have proven to be better than single acute doses of either the synthetic or natural hormone application in all parameters considered ranging from shortest latency and incubation periods, best fertility, hatchability, and survivability, as well as best mean daily growth rate and highest specific growth rate. Besides, the experiment has equally proven that breeding can be successful without the application of hormone provided the female gonad is fully matured and MS222 anesthesia applied during stripping to lessen pain on the fish species.

Recommendation

Though it has been known world-wide that hatchability of *Clarias gariepinus* could be carried out either by using Ovaprim or pituitary hormone, the current research gave credence to the combination therapy of both hormones at one half of the normal dosage of each, proven to be cost effective and gave the best results expected in the fisheries sub-sector to enable the promotion of policies to stop/reduce over-dependence on fish importation.

Alternatively, where hormones are beyond the reach of breeders, eggs could be stripped from matured broodstock that have been properly fed with balanced nutrition and fertilized by mature roe/milt with a caution to stop further stripping upon the slightest taint of blood oozing out.

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Effect of Soil Reaction (Soil pH) on the Rhizosphere Soil, Growth and Yield Performance of Root and Tuber Crops (RTCs) Production: A Review.

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Abstract
Root and tuber crops play a crucial role in global food security, providing a significant source of nutrition and income for millions of people. The rhizosphere, the zone of soil surrounding the plant roots, is a dynamic environment that strongly influences crop growth and development. One critical factor that affects the rhizosphere's functionality is soil reaction, commonly represented by pH levels. This research reviewed the effects of soil pH on the rhizosphere soil, growth, and yield performance of root and tuber crops production. To achieve this, a review of soil pH levels of root and tuber crops was carried out focusing on the key roots and tuber crops such as cassava, sweet potato, potato, yam, and cocoyam etc. Their different soil pH level was observed and noted. It was observed that soil reaction significantly influenced the rhizosphere's physicochemical properties, including nutrient availability, microbial activity, and soil structure. Altered pH levels affected the uptake of essential nutrients, which consequently impacted plant growth and development. Higher or lower pH levels disrupted the balance of nutrients, leading to reduced root and tuber crop yields. Furthermore, soil pH also influenced the rhizosphere's microbial community composition and diversity. Acidic or alkaline conditions affected beneficial microbial populations, such as mycorrhizal fungi and nitrogen-fixing bacteria, leading to potential nutrient deficiencies and reduced plant vigour. In conclusion, maintaining an optimal soil pH in the rhizosphere and soil management practices are essential for maximizing the growth and yield performance of root and tuber crops.
Keywords: Soil pH, Rhizosphere soil, Cassava, Sweet Potato, Potato, Yam, Cocoyam

Introduction

Soil reactions, simply known as soil pH, is a measure of soil acidity or alkalinity which can significantly affect the rhizosphere soil, growth, and yield performance of root and tuber crops. Chemically, it is defined as the $\log_{10}$ hydrogen ions (H^+) in the soil solution. The pH scale ranges from 0 to 14; a pH of 7 is representedas neutral. If pH values are greater than 7, the solution is considered basic or alkaline; if they are below 7, the solution is acidic. It is important to recognize that because the pH scale is in logarithmic units, a change of just a few pH units can induce significant changes in the chemical environment and sensitive biological processes. For example, soil with pH 5 is 10 or 100 times more acidic than soil with pH 6 or 7, respectively (USDA,2011; Brady*et al*,2002). Sources of H^+ ions in soil solution include carbonic acid produced when carbon dioxide (CO_2) from decomposing organic matter, root respiration, and the soil atmosphere is dissolved in the soil water. Other sources of H^+ ions are root release, the reaction of aluminium ions (Al^{+3}) with water, nitrification of ammonium from fertilizers and organic matter mineralization, the reaction of sulfur compounds, rainwater, and acid rain(Karlen *et al*, 2008).

However, the rhizosphere is the region of soil surrounding the root system where various biological, chemical and physical processes take place. That region also influences the soil pH by the release of exudates from the roots which enhance microbial activities, and other physical processes.

It is important to note that before any cropping activities, soil pH has to be tested or crop itself if needed, to ensure optimal growth and quality. Some crops tolerate acid soils whileothers grow well on alkaline soils. The majority of crops grow in soils with pH between 6 and 7.5; few crops prefer, however, acid or alkaline soils.The pH of root and tuber crops such as cassava, sweet potato, yam, cocoyam etc can vary depending on specific cultivars, the soil conditions they are grown in, the maturity of the crop, and the processing methods used(Brady*et al*,2002). Cassava typically has a pH range of 5.5 to 7.5, which falls within the slightly acidic to neutral range. Sweet potatoes have a similar pH range of 5.3 to 6.8, leaning slightly towards the acidic side. Similarly, yams also have a pH range of 5.5 to 6.8, while cocoyam has a pH range of 6 to 6.5. In general, these root and tuber crops prefer slightly acidic to neutral pH levels for optimal growth and development. The pH of the soil is important as it affects nutrient availability, microbial activity, and overall crop health. Most root and tuber crops tend to perform well within the pH range of 5.5 to 6.8, which allows for adequate nutrient uptake and healthy root development(Table 3). The pH values mentioned here may vary slightly depending on specific cultivars, growing conditions, and processing methods (Wang *et al*,2002). Therefore, this paper seeks to review the effects of soil reactions on the rhizosphere soil, growth and yield performance of root and tuber crops production

Factors Influencing pH Values in the Soil

There are a few factors which can influence the pH in the soil which include parental materials of the soil, and the dynamic nature of the land (Smith *et al,* 1996):

1. Inherent Property of Parental Materials

The natural soil pH reflects the combined effects of the soil-forming factors. Weathering and associated chemical reactions release cations (Ca^{+2}, Mg^{+2}, K^{+1}, Na^{+1}) that remain freely in soil solution, form complexes with dissolved organic carbon compounds or are adsorbed at the surface of clay minerals and humus. Over time, the leaching of free cations, basic oxides, and carbonates, as well as the removal of cations by plant uptake and harvesting, and the continuous formation of carbonic acid result in soil acidification. When cations are lost, H^+ ions replace them in the soil solution and on the surface of clay minerals and humus. In very acid soils (where pH is less than 5), aluminium becomes soluble and its ions (Al^{+3}) tend to react with water to produce H^+ ions, which further exacerbate acidification. In arid climates, soil weathering and leaching are less intense, cations accumulate, and OH^- ions offset H^+ ions. As a result, the soil becomes neutral or alkaline. Soils with coarse textures may acidify easily compared to clay soils because they have low organic matter content, a low buffering capacity, a low cation-exchange capacity (poor cation retention), and high rates of water percolation and infiltration. Clay and organic matter in mineral soils act as buffers to resist pH variations. Soil parent material influences soil properties including pH as shown by the contrasting pHs of soil formed in calcareous and granitic materials. The effect of vegetation on soil pH is due partly to the type of humus produced as certain types of humus are soil acidifying. Water erosion removes surface horizons, which can be rich in organic matter, creating a pH gradient along the slope (Smith *et al,* 1996).

2. Dynamic Nature of Agricultural Lands

The conversion of uncultivated land into cultivated soils can result in drastic pH changes after a few periods. These changes are caused by the removal of cations by crops, the acceleration of leaching, the effect of fertilizers and amendments, and the variations in organic matter content and soil buffering capacity. Inorganic amendments (lime and gypsum) and organic amendments rich in cations increase soil pH. Ammonium from organic matter mineralization (nitrification), ammonium-based fertilizers, and sulfur

compounds lower the pH. High rates of water percolation and infiltration increase the leaching of cations and accelerate soil acidification (Neina, 2019).

Soil pH levels and their relationship to soil functions

Soil pH affects the rhizosphere soil's physical, chemical, and biological properties and processes, as well as the growth and yield performance of root and tuber crops. The followings are the effects of soil pH on soil functions (Neina, 2019):

1. Nutrient Availability:

The nutrition, growth, and yields of most crops decrease where pH is low and increase as pH rises to an optimum level. Many crops grow best if pH is close to neutral (pH 6 to 7.5) although a few crops prefer acid or alkaline soils. Soil pH affects the availability and uptake of essential nutrients by root and tuber crops. Different nutrients have varying solubility at different pH levels. For root and tuber crops, maintaining an optimal pH range is crucial for nutrient uptake and utilization. For example, at low pH(acidic conditions), certain essential nutrients like magnesium, phosphorous, calcium and aluminium may become more available and toxic to crops, while at high pH (alkaline conditions), micronutrients like iron, manganese, zinc and copper may become less available. Potatoes prefer slightly acidic soil with a pH range of 5.0 to 6.0, while sweet potatoes prefer slightly acidic to neutral soil with a pH range of 5.5 to 7.0. Imbalance of nutrient availability due to soil pH can lead to nutrient deficiencies or toxicities, resulting in reduced root and tuber crop growth and yield (Neina, 2019).

2. Soil Microbial Activity.

Soil pH directly influences the activity and composition of soil microbial communities in the rhizosphere. Rhizosphere microorganisms play crucial roles in nutrient cycling, organic matter decomposition, biological nitrogen fixation, disease suppression, and production of plant growth-promoting substances. Bacterial populations and activity decline at low pH levels, whereas fungi adapt to a large range of pH (acidic and alkaline). Different microbial species and functional groups have distinct pH preferences. Thereby, most microorganisms have an optimum pH range for survival and function (Table 2). At very acid or alkaline pH levels, organic matter mineralization is slowed down or stopped because of poor microbial activity linked to bacteria. Nitrification and nitrogen fixation are also inhibited by low pH.Acidic soils generally have higher microbial activity, while alkaline soils tend to have lower microbial populations. Certain beneficial microbes, such as mycorrhizal fungi, are more active and efficient in nutrient uptake at specific pH levels. These microbes form symbiotic relationships with crop roots, enhancing nutrient availability and promoting crop growth. Therefore, pH Can influence nutrient availability, diversity, abundance, and activity of beneficial microorganisms in the rhizosphere, indirectly affecting root and tuber crop growth and yield (Neina,2019).

3. Soil Structure and Aeration:

The effects of soil pH on cation availability influence aggregate stability since multivalent cations, such as calcium ions, act as bridges between organic colloids and clays.Soil pH affects soil structure and aggregation, which in turn impacts water infiltration, drainage, and root penetration. Acidic soils with low pH levels can lead to aluminium and manganese toxicity, resulting in poor soil structure and root growth. Alkaline soils, on the other hand, tend to be compacted, reducing aeration and root development. Proper soil pH management helps to maintain a favourable soil structure for root and tuber crop production(Wang *et al*,2002).

4. Root Growth and Development.

Soil pH affects root growth and development in root and tuber crops. Depending on the crop species, different pH ranges may be optimal for root growth. Acidic soils can inhibit root elongation and branching,

leading to reduced nutrient and water uptake. On the other hand, excessive alkalinity can restrict nutrient absorption by reducing root hairs' development. The root systems' overall health and efficiency are crucial for nutrient and water uptake, influencing growth and yield performance.

5. Disease and Pest Occurrence.
Some diseases thrive when the soil is alkaline or acidic.Soil pH can impact the incidence and severity of diseases and pests affecting root and tuber crops. Some pathogens and pests have pH preferences, and changes in soil pH can create favourable conditions for their growth and spread. For example, alkaline soil pH may favour certain root rot pathogens, while acidic soil pH may encourage other diseases. Additionally, changes in soil pH can influence the balance between beneficial and harmful organisms in the rhizosphere, affecting disease suppression mechanisms

6. Tuber Yield and Quality.
Soil reactions have a direct effect on the tuber yield quality in root and tuber crops. Suboptimal pH conditions can result in reduced tuber initiation, size, and overall yield(Table 4). Improper pH levels can also affect tuber quality attributes, such as starch content, tuber colour, and nutritional composition. Additionally, tuber crops like potatoes are susceptible to physiological disorderslike potato scab, which can be influenced by soil pH.

7. Agrochemical Interactions and Mobility of Heavy Metals
The mobility and degradation of herbicides and insecticides, and the solubility of heavy metals are pH dependent. Most pesticides are labelled for specific soil conditions. If soils have a pH outside the allowed range, the pesticides may become ineffective, change to an undesirable form, or may not degrade as expected, which results in problems for the next crop period**.** Many heavy metals become more water soluble under acid conditions and can move downward with water through the soil, and in some cases move to aquifers, surface streams or lakes(Wang *et al*,2002).

Problems with Poor Soil pH Levels and Soil Amendments
Poor pH levels of the soil are associated with the deficiencies of many nutrients, reduction in microbial activity and crop yield, and deterioration of environmental conditions. Therefore, several soil amendments can be used either to increase or reduce soil pH levels (Tlustos, *et al*., 2006, Neina, 2019; Wang *et al*,2002; Smith *et al*, 1996). These steps are taken to improve soil pH to make it more alkaline;
i) Lime; Agricultural lime, also known as calcium carbonate, is commonly used to raise soil pH. It reacts with soil acidity, releasing calcium and raising the pH level. Different types of lime such as calcitic lime or dolomitic lime, can be used depending on the specification needs of the crop.
ii) Wood Ash**:** Wood ash from burned plant material, such as hardwood, can be used as a source of potassium and to raise soil pH. However, it is important to use wood ash in moderation, as excessive use can lead to nutrient imbalances.
iii) Addition of organic residues rich in basic cations, and crop rotation to interrupt the acidifying effect of leguminous crops increase soil pH.

The steps taken to reduce soil pH to make it more acidic are as follows:
i). Sulphur: Elemental sulphur is commonly used to lower soil pH. It reacts with soil moisture and bacteria to produce sulfuric acid, which lowers the pH level. The effectiveness of elemental sulfur depends on soil moisture, temperature and the presence of certain bacteria.
ii). Aluminium Sulfate: Aluminium sulfate is another amendment to lower soil pH. It reacts with soil water to produce aluminium hydroxide and sulfuric acid reducing the pH level. It is important to use aluminium sulfate cautiously, as excessive use can lead to aluminium toxicity.

iii). Application of ammonium-based fertilizers, urea, sulfur/ferrous sulfate, irrigating with acidifying fertilizers, or using acidifying residues (acid moss, pine needles, sawdust) decrease soil pH.

iv). Organic matter: Incorporation of organic matter such as compost or well-rotted manure, can lower soil pH over time. Organic matter releases organic acids as it decomposes which can gradually lower the pH level. Increasing organic matter also increases buffering capacity, meaning that a soil pH will be slower to change with the same action that may more significantly lower the value.Certain soils are more resistant to a drop or rise in pH (buffering capacity). Therefore, the lime requirement, which is the quantity of limestone ($CaCO_3$) required to raise the pH of an acid soil to a desired pH, must be determined specifically for each field before amending the soil.

Measurement of Soil pH

This is an important parameter for determining the acidity or alkalinity of the soil, which directly affects plant growth and nutrient availability. Soil pH Can be measured both in the field and in the laboratory using various methods (Smith *et al*, 1996; Brady *et al*, 2002; Karlen *et al*, 2008).

1. Field measurement:

a) pH Test strips: One of the simple methods is to use pH test strips that change colour based on the pH levels. A soil sample is mixed with distilled water, and the pH test strip is inserted into the mixture. The colour change is then compared to a pH colour chart to determine the soil pH.

b) Portable pH Meters: Portal pH meters are more accurate but also more expensive. A soil sample is mixed with distilled water to create a soil slurry. The pH meter probe is then inserted into the slurry to measure the pH value directly.

c) Soil pH probes: These are similar to portable pH Meters but are more specifically designed for measuring soil pH. Soil pH probes have longer probes to directly measure the pH in the soil without the need for a soil slurry

2. Laboratory Measurement

a) Glass Electrode pH meter: These meters are commonly used in laboratories to directly measured soil pH. A soil sample is mixed with distilled water to create a soil slurry, and the glass electrode pH meter probe is inserted into the slurry to measure pH.

b) Colourimetric Methods: In the laboratory, various colourimetric methods can be used to determine soil pH. The methods often involve using indicators or dyes that change colour with different pH levels. The colour change is then compared to a standard colour chart or measured with a spectrophotometer to determine the soil pH.

c) Titration: A chemical titration method can be used in the laboratory to determine soil pH. A soil sample is mixed with a known concentration of acid or base solution, and then an indicator is added. The soil solution is titrated with a standardized acid or base solution until the indicator changes colour. The amount of acid or base used can be used to calculate the soil pH.

Conclusion

To optimize rhizosphere soil conditions and maximize root and tuber crop production, it is essential to monitor and manage soil pH levels accordingly. Conducting regular soil tests, applying appropriate amendments such as lime or sulfur, and adopting suitable cultural practices can help maintain the desired pH range for optimizing nutrient availability, promoting beneficial microbial activity, supporting root growth, minimizing disease and pest incidence, and ensuring high tuber yield and quality in these crops.

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Table 1. Most Common Classification of Soil pH

Strength of Soil pH	Values
Extremely acid	3.5-4.4
Very strongly acid	4.5-5.0
Strongly acid	5.1-5.5
Moderately acid	5.6-6.0
Slightly Acid	6.1-6.5
Neutral	6.6-7.3
Slightly Alkaline	7.4-7.8
Moderately Alkaline	7.9-8.4
Strongly Alkaline	8.5-9.0

Table 2: Maximum, Minimum and Optimum pH values for microbial groups

Microorganisms	Range	Optimum
Bacteria	5.0-9.0	7
Actinomycetes	6.5-9.5	8
Fungi	2.0-7.0	5
Blue Green Bacteria	6.0-9.0	>7
Protozoa	5.0-8.0	>7

Source; (Smith *et al*, 1996)

Table 3: Soil pH value for Root and Tuber crops

Root and Tuber Crops	Range
Cassava	5.5-7.5
Sweet Potato	5.3-6.8
Potato	6.0-6.8
Cocoyam	5.5-6.5
Yam	5.5-6.8

Source; (Smith *et al*, 1996)

Table 4: Crop Yield Relative to Soil pH

Crops/pH	4.7	5.0	5.7	6.8	7.5
Cassava	34	73	83	100	85
Sweet Potato	65	78	89	100	99
Potato	77	93	99	98	100
Yam	65	79	80	100	93
Cocoyam	31	47	66	100	95

Source; (Smith *et al*, 1996)

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Genetic Diversity of Ugandan Local Chicken Genotypes Inferred from Mitochondrial DNA D-loop Sequences

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Abstract
This study aimed to examine the genetic relationship and structure of the local chicken genotypes in Uganda to serve as an essential component for improvement and conservation strategies. A set of 46 mitochondrial DNA D-loop sequences among 7 chicken strain populations across Uganda was evaluated. The results revealed 5 polymorphic sites defining 7 haplotypes with the dominant (80%) haplotype, UGA01, shared among the populations. Maximum genetic variation was observed among frizzle and rose comb strains which had 2 haplotypes each, followed by the normal feathered which showed the highest (5) haplotypes and crested chickens with 3 haplotypes, however, naked neck, ptilopody, polydactylous chickens were the least diverse with highly conserved sequences. Overall, both haplotype (0.353±0.090) and nucleotide diversities (0.0007±0.0021) revealed a high sequence conversation among the strains, corresponding to a low genetic variation with no genetic structure detected. However, the highest genetic distance observed was between the naked neck / rose comb (0.300) and naked neck/ frizzle (0.300) chickens. A close phylogenetic relationship was observed among the strains suggesting a single founder population adapting to prevailing keeping conditions and high gene flows among the strains. All observed haplotypes were exclusively haplogroup E1 ancestry believed to have its roots in the Indian subcontinent.
Keywords: *Gallus gallus,* chicken genotypes, phylogenetic structure, mutant genes, mtDNA control region, genetic relationship.

1.0 Introduction

Local chickens (*Gallus gallus domesticus*) which dominate the production system as the main poultry group in Uganda (FAO, 2009a) play an important role as part of food security and livelihood of rural economies (FAO, 2009b) as well as urban poor, especially for women and children (Malabo Montpellier Panel, 2020). They are largely preferred for production and consumption by most people (FAO, 2018; Kyarisiima *et al.*, 2004). They are a breed choice due to their adaptability to the local environment and ability to thrive under low input conditions compared to the commercial highly selected hybrid chicken strains, under the prevailing low input production conditions. Nonetheless, the contribution of local chickens to household economies could be better supported by safeguarding their genetic diversity for future sustainability. Access to a wide genetic diversity of animal genetic resources ensures the sustainability of animal production systems and future food security (AU-IBAR, 2019). Local chicken genetic resource constituting an essential part of biodiversity serves as the basis for efficient development in selection and conservation programmes (Groeneveld *et al*., 2010). However, the practical option of ensuring the conservation of chicken genetic diversity for future sustainability is challenged by the continuous introgression of the highly selected commercial chicken lines into the local chicken population (Roothaert *et al*., 2011; Sorensen and Ssewannyana, 2003). This leaves many undiscovered genetic potentialities of the chicken genetic resource at risk of disappearance before even being recognised for any breeding programme. An assessment of the

local chicken breed diversity is thus an eminent requirement for improvement strategies and development to be oriented towards better use of genetic material.

A wide genetic variation reported in the Ugandan local chicken population in many observable forms (Ssewannyana et al., 2008; Yussif et al., 2023) is a consequence of the evolution of alleles by two or more consecutive mutations (Hvilsom *et al*., 2022). Genomic rearrangement in the chicken genome tends to influence observed phenotypic evolution in head features such as wattles, comb (Imsland *et al*., 2012) and tufted crest/ crest feather (Wang *et al*., 2012); feather structure and feather distribution (Chen *et al*., 2008); as well as skeletal variance (Zhang *et al*., 2016). The phenotypic evolution enables adaptation to various environmental conditions through a complex pattern of interactions and signals. For instance, feather mutations such as frizzle, nakedness, slow feathering, naked neck, tufted crest and by extension skin colouration, fibromelanosis genes (Jin *et al*., 2016; Gunnarsson, 2009) are useful traits in a hot environment and are adapted for the outdoor management system (Fernandes *et al*., 2023; Wang *et al*., 2012). Polydactyl and ptilopody (feathered shank) phenotypes tend to have a high growth rate and egg performance potential (Dahloum *et al*., 2016; Mensah, 2016; Adomako, 2009). The local chickens segregate for these major genes to adapt to the prevailing outdoor management system in hot tropical climates. It could therefore be assumed that they contain alleles that contribute to altered morphology in those carrying them which is relevant to their adaptations peculiar to their specific management and environmental circumstances. Hence, using molecular markers, genetic assessment of the diversity among the chickens showing genes that characterise major genotypes with known adaptive values would be better understood for purposes such as conserving superior genotypes and developing efficient breeding programmes. Such information is necessary as a vital complement to the evaluation of phenotypic diversity and the production environment (FAO, 2012; FAO, 2011).

The mitochondrial DNA (mtDNA) control region has been used as a marker to study genetic diversity in chicken populations because they are highly polymorphic with the absence of recombination. Thus, a powerful tool to measure genetic diversity indices. Sequence variation analysis of Displacement (D)-loop is sufficiently supported by the complete mtDNA genome (Miao *et al*., 2013) and could provide a valuable description of the population structure, demographic history, geographic origins and adaptation (Lan *et al*., 2015). The population structure of chicken has been explored using mtDNA sequences of 3 Nigerian genotypes (Ikpeme *et al*., 2021) and Egyptian fayoumi chicken (Eltanany and Hemeda, 2016). It is widely accepted that detailed molecular genetic variation data on breed diversity are essential for effective selection and crossbreeding management of livestock (AU-IBAR, 2019). Therefore, this study assessed the genetic relationship and structure of Ugandan chickens carrying genes that characterise major genotypes/ strains by variation analysis of the mtDNA D-loop sequence. This was aimed at gaining an understanding of the molecular diversity of the chicken genetic resources of Uganda to guide genetic improvement and conservation strategies.

2.0 Materials and Methods

2.1 Study Design, Blood Sampling and Molecular Laboratory Procedures

Whole blood samples were obtained from a total of 46 local chickens segregating for genes that characterise major genotypes: Rose comb, crested chicken, ptilopody, naked neck, frizzle feather, normal feather and polydactyl chickens in local-chicken-keeping households across Uganda (**Fig. 1**). Approximately 1 ml of blood from the ulnar wing vein was collected into labelled 2 mL microtubes one containing 1 mL absolute ethanol following the protocol for genetic-based assays by Owen (2011) and stored at -20 ℃.

Genomic DNA was isolated from 10 µl blood following the Bench Protocol (Spin-Column Protocol) designed for nucleated erythrocytes (QIAGEN, 2020). Visual estimation of the genomic DNA profile was done in a UV Transilluminator documentation system (G-BOX, SYNGENE). The fragment, 800 bp of mtDNA D-loop region was amplified, using previously reported primer pair: AV1F2: 5'-

AGGACTACGGCTTGAAAAGC-3' / CR1b: 5'-CCATACACGCAAACCGTCTC-3' (Mwacharo *et al*., 2011), in a polymerase chain reaction (PCR). The amplification was performed for each DNA isolate in a 25 µl reaction volume of mix containing; 2 µl (30 ng) genomic DNA, 0.5 (10 pMol/ µl) each of the primer pair and 5 µl of 5x FIREPol® Master Mix Ready to Load (Solis BioDyne)(Solis BioDyne, 2020). The final reaction volume was adjusted with ultrapure PCR-grade water and the amplification processes proceeded on the Applied Biosystems® Veriti® 96-Well Thermal Cycler (Thermo Fisher Scientific). Thermal cycling conditions comprised initial denaturation 95℃ (1min), denaturation 95℃ (15s), annealing 58℃ (1min), elongation 72℃ (1min), 35 cycles final elongation 72℃ (10min), and held at 4℃ until remove from. The PCR products were verified by Ethidium bromide (EtBr)-stained gel (1.5% agarose in 100 ml 1x TAE buffer, 150V, 40 mins) electrophoresis and detected under UV light.

Amplified DNA fragment size was estimated through size comparison with a 1 kb DNA ladder ready-to-load (Solis BioDyne) loaded alongside. Purification of the amplicons was done using the PureLink® PCR Purification Kit (Thermo Fisher Scientific Inc.). Sanger sequencing reactions with each of the primer pairs were performed on the *Standard-seq* platform of Macrogen Europe B.V. (https://dna.macrogen-europe.com).

Figure 5: Geographic location of the Ugandan chicken strains and their phenotypic appearance.

2.2 DNA Sequence Alignment and Analyses

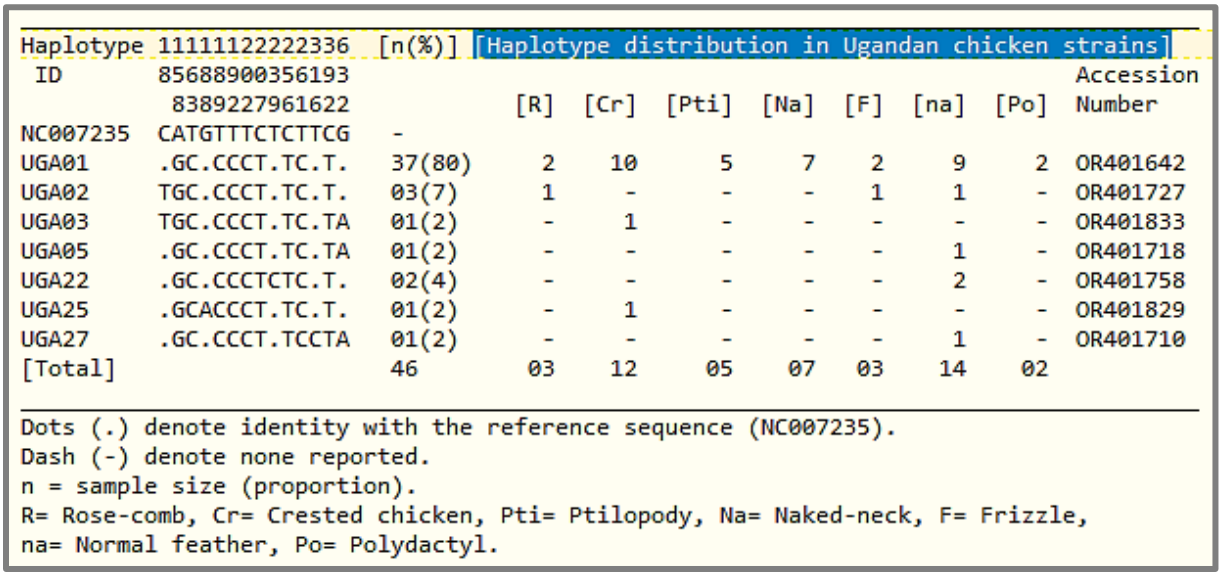
DNA Sequences were subjected to chromatogram quality control in FinchTV v1.4.0 software (Geospiza, Inc), and assembled in BioEdit v7.2.5 (Hall, 1999). Multiple Sequence Alignment was conducted using the MUSCLE algorithm in MEGA v11 (Tamura *et al.*, 2021). Alignments were refined manually to set nucleotide base pairs at 658 bp (651 bp, excluding sites with gaps). Sequence polymorphism parameters and distribution of haplotypes were measured using DnaSP v6.12.03 (Rozas *et al.*, 2017), with one outgroup (GenBank accession number NC_007235). Within-population diversity parameters; haplotype diversity (*Hd*), with corresponding nucleotide diversity (π), and average number of nucleotide differences between haplotypes (*k*) were tested in Arlequin version 3.5.2.2 (Excoffier *et al.*, 2005). Analysis of molecular variance (AMOVA) to partition the total genetic variance into components was conducted in Arlequin 3.5.2.2 software with 1000 bootstrap replications (Excoffier *et al.*, 2005). Genetic distance among the 7 strains was measured by the pairwise F_{ST} value and the statistical tests of the pairwise F_{ST} values were estimated by permutation analysis using 1000 replications in Arlequin 3.5.2.2 software. The evolutionary relationships among the strain haplotypes were visualized by the construction of a median-joining network (Bandelt *et al.*, 1999) using PopArt 1.7 7 (http://popart.otago.ac.nz) and neighbour-joining tree based on p-distance method (Nei and Kumar, 2000) was conducted in MEGA v11 (Tamura *et al*., 2021). All observed haplotype sequences are part of sequences deposited at the NCBI Genbank with accession numbers OR401589-OR401932.

3.0 Results and Discussion

3.1 Sequence Polymorphism, Haplotype Distribution and Diversity

The mtDNA D-loop sequence polymorphism and distribution of haplotypes in the chicken strains are presented in **Fig. 2**. A total of 5 segregating sites defined 7 unique haplotypes of which the dominant (80%) haplotype detected among all the strains is UGA01. Haplotype UGA02 was shared by rose comb, frizzle and normal feather strains. The rest of the haplotypes were singletons detected only in one particular strain. Four segregating sites were detected in normal feather chicken, 1 in frizzle feather, 3 in crested chicken, and 1 in rose comb strain. However, the D-loop sequences of ptilopody, polydactyl and naked neck strain were highly conserved with no polymorphic site detected and thus constituted 1 haplotype each. Sequence polymorphism indices presented in **Table 1** showed that 5 haplotypes were found in normal feather strains with haplotype diversity (*Hd*) of 0.593±0.144. Frizzle feather and rose comb strains had 2 haplotypes each and showed the highest number of haplotype diversities, 0.667±0.314 each. Crested chickens had 3 haplotypes and the lowest haplotype diversity, 0.318±0.164. Nucleotide diversity was highest in normal feather (0.00125±0.00040) followed by frizzle feather (0.00102±0.00048), rose comb (0.00102±0.00048) and crested chicken (0.00077±0.00044). Overall, both the haplotype and nucleotide diversity suggested low genetic variation among the Ugandan chicken strains as indicated by the low haplotype and nucleotide diversity, 0.353±0.090 and 0.0007±0.0021, respectively. The average number of nucleotides differences among sequences in the population was 0.4589 whereas the overall Tajima's D estimate, -1.4472 was not statistically significant ($\rho > 0.10$), hence, consistent with a population at a in neutral mutation. This suggests that the genetic variations were unaffected by natural selection (Hvilsom *et al*., 2022). These results further suggest a much lower selection pressure on the ptilopody, polydactyl, and naked neck strains compared to the normal feather, frizzle feather, crested-chicken and rose-comb. Polydactyl allele frequency has been reported to be lower than their expected Mendelian proportions in Ugandan indigenous chickens (Yussif *et al*., 2023). Naked neck chickens are traditionally deemed undesired for keeping (Desta *et al*., 2013) and cannot be served to a guest for consumption or fetch a good market price, thus, negatively selected (Yussif *et al*., 2023). Meanwhile, the influence of the naked neck gene (*Na*) on increasing growth performance under tropical conditions is well resolved (Fernandes *et al*., 2023; Fathi *et al*., 2013; Adomako, 2009). The higher degree of diversity in the normal feather, frizzle feather, crested chicken and rose comb strain than the other strains reflect their preference, perhaps as fancy chickens and/or ability to fetch a premium price

in Uganda. A high genetic variation could increase the ability of a population to evolve and hence, increase their chances of survival under the constant environmental changes (Hvilsom *et al*., 2022). The estimates of haplotype diversity observed in this study are similar to those reported for normal feather and frizzle feather chickens in Nigeria based on the mitochondrial *Cyt b* sequences (Ikpeme *et al*., 2021).



Haplotype 11111122222336	[n(%)]	[Haplotype distribution in Ugandan chicken strains]							
ID 85688900356193									Accession
8389227961622		[R]	[Cr]	[Pti]	[Na]	[F]	[na]	[Po]	Number
NC007235 CATGTTTCTCTTCG	-								
UGA01 .GC.CCCT.TC.T.	37(80)	2	10	5	7	2	9	2	OR401642
UGA02 TGC.CCCT.TC.T.	03(7)	1	-	-	-	1	1	-	OR401727
UGA03 TGC.CCCT.TC.TA	01(2)	-	1	-	-	-	-	-	OR401833
UGA05 .GC.CCCT.TC.TA	01(2)	-	-	-	-	-	1	-	OR401718
UGA22 .GC.CCCTCTC.T.	02(4)	-	-	-	-	-	2	-	OR401758
UGA25 .GCACCCT.TC.T.	01(2)	-	1	-	-	-	-	-	OR401829
UGA27 .GC.CCCT.TCCTA	01(2)	-	-	-	-	-	1	-	OR401710
[Total]	46	03	12	05	07	03	14	02	

Dots (.) denote identity with the reference sequence (NC007235).
Dash (-) denote none reported.
n = sample size (proportion).
R= Rose-comb, Cr= Crested chicken, Pti= Ptilopody, Na= Naked-neck, F= Frizzle,
na= Normal feather, Po= Polydactyl.

Figure 6: **S**equence polymorphism of the 7 haplotypes observed in the D-loop region of Ugandan chicken strains alongside their frequencies and distribution. Mutations are scored relative to the reference sequence (GenBank accession No. NC007235). Vertically oriented numbers indicate the site and the sequences shown are only the variable sites.

Table 8: Nucleotide polymorphism indices of the Ugandan chicken strains

Population	n	V	Pi	S	H	*Hd*±SD	π±SD	*k*
Normal feather (na)	14	4	2	2	5	0.593±0.144	0.00125±0.00040	0.8132
Frizzle (F)	3	1	0	1	2	0.667±0.314	0.00102±0.00048	0.6667
Crested chicken (Cr)	12	3	0	3	3	0.318±0.164	0.00077±0.00044	0.5000
Rose comb (R)	3	1	0	1	2	0.667±0.314	0.00102±0.00048	0.6667
Ptilopody (Pti)	5	0	0	0	1	0	0	0
Polydactyl (Po)	2	0	0	0	1	0	0	0
Naked neck (Na)	7	0	0	0	1	0	0	0
Overall	46	5	3	2	7	0.353±0.090	0.0007±0.0021	0.4589

Hd = Haplotype diversity, π = Nucleotide diversity, n = Number of sequences, V = Polymorphic sites, Pi = Parsimony-informative site, S = Singleton site, H = Number of haplotypes, SD = standard deviation, *k* = average number of nucleotide differences. Tajima's = -1.4717 (ρ = 0.9352)

3.2 Genetic structure analyses

Analysis of molecular variance to partition the genetic-variance components did not detect any genetic structure with clear differences among the Ugandan chicken strain populations as indicated by the negative variance component among population. All the genetic variation was attributed to variation within population (**Table 2**). The high within population diversity detected reflects the low nucleotide diversity with a low number of polymorphic loci (0–5) (**Table 1**); and it is consistent with the wide phenotypic variation reported among Ugandan indigenous chickens (Yussif *et al*., 2023; Ssewannyana *et al*., 2008).

The pairwise F_{ST} values for genetic distance among populations are presented in **Table 3.** The highest genetic distance was between the naked neck / rose comb strain (0.300) and naked neck/ frizzle chicken (0.300) populations. Hence, naked neck chickens were most distantly related to frizzle and rose comb strains, in agreement with reports elsewhere (Abou-Emera *et al*., 2017; M. Fathi *et al*., 2018; Ikpeme *et al*., 2021). This high genetic distance among these strains may be due to the numerous genes influencing the naked neck and rose comb genotypes (Imsland *et al*., 2012) as well as the several modifying genes associated with frizzle feather (Hutt, 1949). Structural rearrangements of alleles have been reported to contribute to morphogenesis in natural populations (Imsland *et al*., 2012). The low and non-significant negative F_{ST} obtained indicates that these populations were of similar genetic structure with high gene flow among them; similar to what was observed among Nigerian chicken strains (Ikpeme *et al*., 2021).

Table 9: **Analysis of Molecular Variance (AMOVA): Ugandan chicken strains**

Source of variation	d.f.	Variance components	Variation (%)	Fixation Index, F_{ST}*	*ρ*-value
Among populations	6	-0.01326	-5.84	-0.0584	0.8192
Within populations	39	0.24023	105.84		
Total	45	0.22697	100		

*The sum of variation groups divided by total variation. d.f.= Degrees of freedom.
Negative values of variance occur in the absence of genetic structure at a given level

Table 10: **Population pairwise F_{ST} values (Distance method: Pairwise difference)**

Chicken strain	F_{ST} values / *ρ*-values						
	R	Cr	Pti	Na	F	na	Po
Rose comb (R)		0.487	0.376	0.297	0.999	0.776	0.999
Crested chicken (Cr)	-0.067		0.999	0.999	0.520	0.760	0.999
Ptilopody (Pti)	0.189	-0.093		0.999	0.389	0.694	0.999
Naked neck (Na)	0.300	-0.051	0.000		0.245	0.613	0.999
Frizzle (F)	-0.500	-0.067	0.189	0.300		0.749	0.999
Normal feathered (na)	-0.062	-0.024	-0.057	-0.016	-0.062		0.999
Polydactyl (Po)	-0.200	-0.327	0.000	0.000	-0.200	-0.279	

Above diagonal: ρ-values of pairwise F_{ST} values.
Below diagonal: pairwise F_{ST} estimates between populations

3.3 Phylogenetic relationship among the Ugandan chicken strains

Results of the phylogenetic analyses through median-joining and neighbour-joining trees revealed similar topologies in **Fig. 3**, showing the close genetic relationship among the populations. The median-joining network showed a conformation which radiated from the predominant haplotype, UGA01, suggesting a root haplotype in Uganda composed of individuals from the different genotypic strains (**Fig. 3A**). Three divergent subclusters were observed among the chicken strain population inferred by the neighbour-joining tree, all of which showed exclusive affinity to the haplogroup E1 ancestral lineage (**Fig. 3B**) of Miao *et al*. (2013) mtDNA classification. This lineage forms the most widely distributed domestic chicken ancestry globally with its centre of diversity in the Indian subcontinent (Miao *et al*., 2013). Individuals of all the chicken strains were found in one big subcluster which was solely of the haplotype UGA01. However, individual normal feathered, frizzle and rose comb strains, although among the haplotype UGA01, were found in different subcluster with one crested chicken of haplotype UGA05 and some individual normal feathered chickens of multiple haplotypes, UGA25 and UGA27. The normal feathered chicken was observed in all 3 subclusters, suggesting an original ancestral population that was likely established first in Uganda and then expanded to other strains by one or two mutations. It is maintained that past climatic and

geographic events could affect patterns of sequence divergence for adaptive survival (Hvilsom *et al*., 2022). The result of chicken strain individuals clustering into one major group was detected in Nigerian local chicken genotypes, where normal feathered, frizzle and naked neck chickens were in the same cluster (Ikpeme *et al*., 2021).

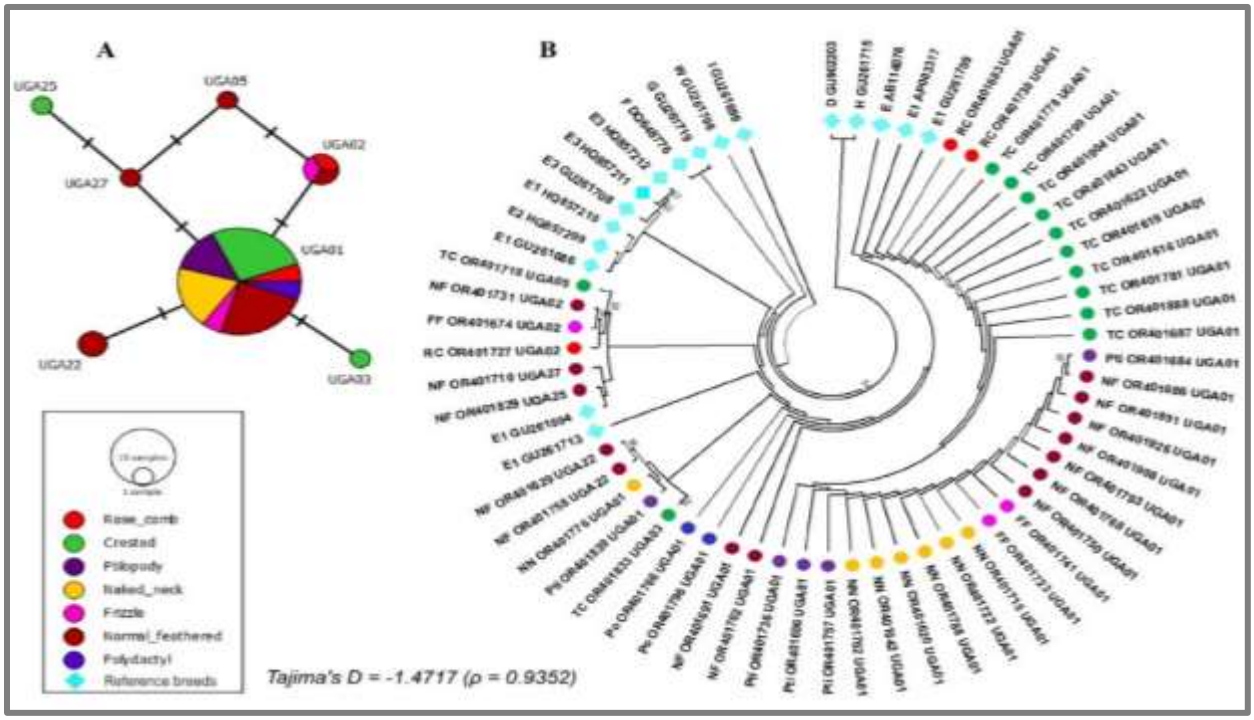


Figure 7: Phylogenetic relationship among the Ugandan chicken strains: **(A)** A Median-joining network profile of the mtDNA D-loop haplotypes. The area of the circles is proportional to haplotype frequency and the hatch marks on the line correspond to mutational positions connecting haplotypes. **(B)** A Neighbour-Joining tree showing the phylogenetic relationships among the strains and 17 reference breeds from Miao *et al*. (2013) mtDNA haplogroup classification.

4.0 Conclusion

In this study, the genetic relationship and structure of Ugandan chickens carrying genes that characterise major genotypes were assessed by variation analysis of partial sequences of the mtDNA control region. It was revealed that a higher genetic diversity exists among frizzle, rose comb, normal feathered and crested chickens compared to naked neck, ptilopody and polydactylous strains with a great amount of within-strain population genetic variations. However, genetic structure was not detected among populations with the different strains distributed in one dominant haplotype (UGA01) of haplogroup E1 mtDNA lineage originating from the Indian subcontinent. This confirms a close relationship among all strains, likely as a consequence of a founder effect and high gene flows. This study forms the initial investigation using a molecular marker in these chicken populations in Uganda; and may be useful in developing wider in-depth research strategies for improved breeding and conservation of valuable indigenous chicken genetic resources.

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Multi-location Evaluation of Advanced Mungbean Lines under Different Climatic Conditions in Uganda

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Abstract
Mungbean [*Vigna radiata* (L.) R. Wilczek var. *radiata],* or greengram is a popular crop especially in the drought prone areas of East Africa. It is rich in micronutrients and protein thus can help ameliorate malnutrition if incorporated in diets. Yield stability across target population of environments is a critical trait for mungbean breeding in the region. In this study, i) we examined the influence of climatic covariables on G X E for grain yield among mungbean genotypes ii) determined yield stability of mungbean genotypes across environments in Uganda. Field trials comprising of 40 mungbean genotypes were established in 9 environments in Uganda in an alpha lattice layout. Our findings showed that radiation and minimum relative humidity were the key factors responsible for G x E on mungbean yield. These contributed 40% of the G X E. Four mungbean genotypes; AVMU2021, AVMU2006, UMG2017-211 and UMG2017-263 exhibited both stability and adaptability across the test environments. Additional evaluation will be conducted for their potential release as varieties and also inclusion in the mungbean breeding program as parents.
Keywords: Factorial regression; Genetic variation; Stability; G X E; Greengram; Yield

Introduction

Mungbean (*Vigna radiata* (L.) also known as greengram is a key food and cash crop in several parts of the world especially in south Asia and east Africa. In Uganda, the crop is popular I the eastern and northern parts of the country especially among small-scale farmers (Mbeyagala et al., 2016, 2017). Mungbean has short growing cycle, requires less inputs compared to other crops, is tolerant to drought and also improves soil fertility through biological nitrogen fixation making it suitable for low input production systems (Nair and Schreinemachers, 2020). In terms of nutritional quality, mungbean grain is a rich source of essential nutrients e.g proteins (up to 31%), iron (>87 mg/kg) and zinc (up to 62mg/kg) (Nair et al. 2015; Anwar et al. 2007). The grain contains less flatulence causing factors compared to other legumes. This makes it an ideal food for children, the elderly and the convalescing (Adsule et al. 1986). Mungbean starch is also highly digestible with a high glycaemic index and thus more suitable for malnourished patients (Sandhu and Lim, 2008). Mungbean is consumed in a several ways as whole grain or as split grain, processed into flour or as bean/ vegetable sprouts (Nair and Schreinemachers, 2020).

Given its rich nutritional profile, mungbean has the potential to alleviate nutritional disorders especially in developing countries like Uganda where consumption of fortified foods and food supplements is still out of reach for the most vulnerable populations (Dahiya et al 2015). Iron deficiency is the most prevalent form

of nutritional disorder affecting mainly the poorest and vulnerable populations in resource constrained environments in developing countries (Stevens et al. 2022). For instance, in 2019, incidence of iron deficiency anaemia in east Africa was 53%, 31% and 39% among children (<5 years), non-pregnant women, and pregnant women, respectively (Stevens et al. 2022). A food based approach that promotes cultivation and consumption of popular nutrient rich crops/varieties is a more cost effective and sustainable approach (Dahiya et al 2015). Identification of best varieties/lines/cultivars for different locations or environments and which are also stable across environments requires the establishment of multi-environmental trials (METs). Cultivars tested in METs often vary in response from one environment to another due to genotype by environment interaction (G X E) phenomenon (Malosetti et al., 2013; Vargas et al. 1999). Several models such as ANOVA, regression on the mean model and AMMI have been used to describe G X E (Malosetti et al., 2013; Purchase et al. 2000; Purchase, 1997). However, these models do not make of use of explicit external environmental information but only rely on the phenotypic response variable of interest such as yield (Malosetti et al., 2013; Vargas et al. 1999). On the other hand, factorial regression model makes use of external environmental information directly to determine which variables influence G X E. In this study, i) we describe the influence of climatic variables on G X E for grain yield among mungbean genotypes using factorial regression model, ii) determine yield stability of mungbean genotypes across environments in Uganda.

Methods and Methods

Genetic Material

Forty mungbean genotypes comprising of 25 advanced lines from the World Vegetable Center (WorldVeg), formerly known as Asian Vegetable Research and Development Center (AVRDC), 13 accessions from the genebank at NaSARRI and two (2) released varieties (NAROGRAM1 and NAROGRAM 2) were evaluated (Table 1). The released varieties served as checks.

Test Environments

Multi-location field trials were established at five sites in Uganda; Serere at NaSARRI (2020B, 2021A and 2021B seasons), Arua at Abi Zonal Agricultural Research and Development Institute (AbiZARDI) during 2021A and 2021B seasons, Lira at Ngetta Zonal Agricultural Research and Development Institute (NgettaZARDI), Mayuge at Ikulwe satellite station (2021B season) and Kitgum at Kitgum District Farm Institute (DFI) during 2021B season. Each location and season combination constituted a different environment, hence there was a total of nine test environments. The coordinates of trial sites are as provided in Table 2.

Experimental Layout

Field trials were established in an alpha lattice design replicated twice in each environment. Designs were generated each season using Breeding Management System (BMS). The genotypes were planted in two (2) rows each with a length of two (2) metres long with a spacing of 0.5m x 0.1m. At each trial site, planting, weeding, thinning, pest management and harvesting were carried out manually. Pods harvested from each plot were sun-dried before manual threshing. Threshed grain was dried to a moisture content of 13% before determining plot grain weight.

Data Collection: Genotypes

Observations were recorded on several traits (Table 3) during the trials such as on number of days to 50% flowering, number of days to maturity, plant height, pod number, pod length, pod and seed weight, number of seeds per plant, plot pod weight, plot seed weight and seed size (100 seed weight). Plot seed weight was used to compute yield per ha. A description of the trait measured and corresponding stage of observation during field experiments is provided in Table 3 below.

Data Collection: Weather Variables

Records of weather observations during growing period were obtained from Automatic Weather Stations (ADCON Model) installed by Uganda National Meteorological Authority (UNMA) at or near the trial sites. The covariables recorded were average temperature, minimum temperature, maximum temperature, rainfall, radiation, average % relative humidity, minimum % relative humidity, maximum % relative humidity, average wind speed and maximum wind speed.

Data Analysis

Stability and Factorial regression analysis were carried using GEA-R (Genotype by Environment Analysis with R) Software version 4.1 (Pacheco et al., 2015). The climatic covariables listed above were in factorial regression analysis to guide in choosing which of the covariates provides greater proportion of the variability in G X E, explained by differential genotype sensitivity to explicit external environmental variables and have the advantage that hypotheses about the influence of those external variables on G X E of yield can be statistically tested. The factorial regression model for which G X E incorporates environmental covariates used as follows;

$$Y_{ij} = \mu + g_i + e_j + \sum_{g=1}^{H} Z_{ih}\,\zeta_{jh} + \varepsilon_{ij}$$

Where Y_{ij} is the yield of the i-th genotype (i=1,...,I) in the j-th environment (j=1,...,J); μ is the grand mean; g_i and e_j are the genotype and environment deviations from the grand mean, respectively; z_{ih} are the environmental covariates; ζ_{jh} are the genotype factor; H (H < J) is the number of environmental covariates and ε_{ij} is the error term. Summary statistics were computed using doBy R package version 4.6.18 (Højsgaard and Halekoh, 2023).

Results

Performance of genotype for various traits

Number of days to 50% flowering ranged from 37 days (AVMU2015) to 49 days (UMG2017-235) with a mean of 40 days (Table 4). Average number of days to physiological maturity was 66 days with AVMU2001 as the earliest maturing genotype (60 days) and UMG2017-235 as the late maturing genotype (77 days). Average plant height was 27cm with the shortest genotype being AVMU2005 (21cm) and the tallest genotype (UMG2017-139) with a plant height of 40.2cm. On average, each of the genotypes had about 13 pods per plant with AVMU2002 having the lowest number of pods per plant (9pods) while UMG2017-222 had the highest number of pods per plant (20 pods). The genotype with the shortest pods was AVMU2001 (6.3cm) while UMG2017-139 had the longest pods (8.4cm). Genotype AVMU2022 had the lowest number of seeds per pod while UMG2017-139 had the highest number of seeds/pod. In terms of grain yield, the lowest yield genotype was AVMU2025 (681Kg/Ha) while the highest yielder was UMG2017-242 (1394kg/ha). The evaluated genotypes were generally large seeded (with 6.3g/100 seeds). UMG2017-97 had the smallest seed size (4.7g/100 seeds) while AVMU2020, AVMU2021 and UMG2017-211 had the largest seed size (7.4g/100seeds) [Table 4].

Across test environments, genotypes flowered earlier in Serere and later in Arua and Mayuge. Similarly, genotypes attained physiological maturity earlier in Serere compared to Arua (Table 5). on the other hand, yield/ha was highest in Mayuge, Lira and Serere but lowest in Kitgum and Arua. Results of factorial regression model fitted (Table 6) to the mungbean yield data showed that GEI was mainly explained by differential genotype sensitivities to radiation (F=2.36, $P\leq$0.00002) and minimum % relative humidity (F=1.61, P<0.015) during the growing period. Together, radiation and minimum % relative humidity, accounted for up to explained 40% of GEI with 76 degrees of freedom.

Yield stability analysis identified four genotypes that were both stable and adaptable (Fig. 1). These were; AVMU2021, AVMU2006, UMG2017-211, and UMG2017-263. Genotypes that only showed adaptability were; AVMU2018, AVMU2021, NAROGRAM2, AVMU2003, UMG2017-211, UMG2017-263, UMG2017-15, AVMU2006 and AVMU2007.

Discussion

In this study, variation in yield due to GEI was 40% and this was largely attributed to radiation and minimum relative humidity. This GEI variation is much higher than past findings reported by Imrie and Shanmugasundaram (1987). A recent study by Karimi et a. (2019) however, reported rainfall (distribution and amount) and temperature as key climatic parameters responsible for the variation. Other environmental variables such as soil characteristics (type, texture, fertility levels) contribute to differences in genotype performance (Karimi et al. 2019; Nair et al. 2015). However, these were not investigated in this study. This highlights the need to consider G X E in selection of suitable/potential genotypes. Variation was observed among genotypes across environments. Taller mungbean plants are often associated with yield advantage as such plants tend to have more branches and more pods that those with a short stature (Imrie and Shanmugasundaram, 1987). Additionally, mungbean genotypes with a high plant stature are preferred for both manual and mechanized harvesting, a key determinant for variety adoption (Karimi et al. 2019). On the basis of days to flowering and maturity, most of the genotypes tested were early maturing ($\leq$65 days). Similar results were reported by Yimram et al. (2009). Such genotypes are ideal for double cropping systems [2 seasons/year] (Mbeyagala et al. 2017; Yimram et al., 2009). Early maturity has also been reported as a key trait considered by farmers as a basis for mungbean variety selection in Kenya (Karimi et al 2019). Grain/seed size is another key trait for farmers in east Africa with large or bold seeded varieties preferred to small seeded types (<5g/100 seeds) (Mbeyagala et al., 2017). In this study, majority of the genotypes tested were large seeded, with average seed size of 6.3g/100 seeds.

This study identified four genotypes; AVMU2021, AVMU2006, UMG2017-211, and UMG2017-263 that were both adaptable and stable in yield across test environments. Information on stability and adaptability of performance of genotypes or breeding materials can aid farmers in making decisions about adoption and also for the variety release process by breeders (Ceccarelli, 2012).

Conclusion

In this study, radiation and minimum % relative humidity were the significant contributors to GEI for grain yield among mungbean genotypes. Among the genotypes tested, four (AVMU2001, AVMU2006, UMG2017-211, UMG2017-263) were both adaptable and stable in yield performance across environments. These genotypes were also large seeded, a key farmer preferred trait. Further evaluation of these potential genotypes geared towards release as varieties and inclusion in the breeding program as parents is recommended.

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Table 11: List of mungbean genotypes used in the study

No.	Line code	Seed Lustre	Seed Color	Source
1	AVMU2001	Shiny	Pale green	WorldVeg
2	AVMU2002	Shiny	Pale green	WorldVeg
3	AVMU2003	Dull	Green	WorldVeg
4	AVMU2004	Shiny	Pale green	WorldVeg
5	AVMU2005	Shiny	Pale green	WorldVeg

6	AVMU2006	Dull	Green	WorldVeg
7	AVMU2007	Dull	Green	WorldVeg
8	AVMU2008	Dull	Green	WorldVeg
9	AVMU2009	Shiny	Pale green	WorldVeg
10	AVMU2010	Shiny	Pale green	WorldVeg
11	AVMU2011	Shiny	Pale green	WorldVeg
12	AVMU2012	Dull	Green	WorldVeg
13	AVMU2013	Dull	Green	WorldVeg
14	AVMU2014	Shiny	Pale green	WorldVeg
15	AVMU2015	Dull	Green	WorldVeg
16	AVMU2016	Dull	Pale green	WorldVeg
17	AVMU2017	Dull	Green	WorldVeg
18	AVMU2018	Dull	Green	WorldVeg
19	AVMU2019	Dull	Green	WorldVeg
20	AVMU2020	Dull	Green	WorldVeg
21	AVMU2021	Dull	Green	WorldVeg
22	AVMU2022	Dull	Brown	WorldVeg
23	AVMU2023	Shiny	Pale green	WorldVeg
24	AVMU2024	Dull	Green	WorldVeg
25	AVMU2025	Shiny	Pale green	WorldVeg
26	UMG2017-222	Shiny	Pale green	NaSARRI
27	NAROGRAM1	Dull	Green	NaSARRI
28	NAROGRAM2	Shiny	Pale green	NaSARRI
29	UMG2017-97	Dull	Green	NaSARRI
*30	UMG2917-156	Dull	Green	NaSARRI
31	UMG2017-197	Dull	Green	NaSARRI
32	UMG2017-35	Dull	Green	NaSARRI
33	UMG2017-211	Dull	Green	NaSARRI
34	UMG2017-263	Dull	Green	NaSARRI
35	UMG2017-306	Shiny	Pale green	NaSARRI
36	UMG2017-23	Dull	Green	NaSARRI
37	UMG2017-15	Dull	Green	NaSARRI
38	UMG2017-235	Dull	Green	NaSARRI
39	UMG2017-242	Dull	Green	NaSARRI
40	UMG2017-139	Dull	Green	NaSARRI

*UMG2917-156 was not included in analysis because poor establishment across environments

Table 12: Location coordinates of trial sites

Location	Latitude	Longitude	Altitude (m)
Serere	1.536744N	33.447120E	1140
Lira	2.297487N	32.914027E	1084

Arua	3.078586N	30.947898E	1198
Mayuge	0.438806 N	33.476585E	1192
Kitgum	3.290085 N	32.887770E	941

Table 13: Description of traits measured in field evaluation of mungbean genotypes

Trait	Code	Description	Stage
Days to 50% flowering	DFF	Number of days from planting to when at least 50% of the plants in a plot have at least one unopened flower	Flowering
Plant Height (cm)	PH	Recorded on 5 randomly sampled plants per plot	Physiological maturity
Days to Maturity	DM	Days from sowing to when 90% of pods in a plot change color (to black or brown)	Physiological maturity
Pods per plant	NP	Mean number of pods from 5 random plants per plot	After harvest
Pod length (cm)	PL	Mean lengths measured from five randomly selected pods from each plant (up to 5 plants)	After harvest
Pod yield per plant (g)	PY	Mean weight for all pods measured from five randomly selected plants	After harvest
Number of Seeds per pod	NS	Mean number of seeds counted from five randomly selected pods from each plant (up to 5 plants)	After harvest
Seed yield per plant (g)	SY	Mean weight for all seeds measured from five randomly selected plants	After threshing
Plot seed weight (g)	PSW	Weight of all seeds from all plants in a plot	After threshing
Yield (kg)	SYHa	Weight of seeds per ha	After threshing
100 Seed weight (g)	SW_100	Weight of 100 randomly selected seeds	After threshing

Source: IBPGR. 1980.

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Prevalence of Hydatid Cysts in Sheep and Goats Slaughtered in Selected Abattoirs in Northern Nigeria

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Abstract
This study on prevalence and fertility of hydatid cysts of *Echinococcus granulosus* in Sheep and Goats slaughtered n at selected abattoirs in Kaduna, Kogi, Plateau, Gombe States and the Federal Capital Territory, Northern Nigeria was conducted between February to December, 2022. Gross examination was carried out by visual inspection and palpation of lungs and liver of 906 Sheep and 1,924 Goats. Out of a total of 2,830 ruminants, 262(14.3%) harboured one or more cysts in liver (6.7%), lungs (3.6%) or in liver and lungs (1.0%). Highest prevalence was recorded in Sheep (9.6%) than Goats (9.1%). Difference in prevalence of cysts among Sheep and Goats was not statistically significant ($p>0.05$). There was association of prevalence of cysts with Sheep (OR=1.062, 95% CI=0.810-1.391). Prevalence of cysts based on location revealed higher prevalence in 38.9% of sheep and 16.0% of goats slaughtered in selected abattoirs in Kaduna State followed by 23.0% sheep and 16.0% goats slaughtered in Gombe State, least prevalence of 3.0% each was obtained in Goats slaughtered in Plateau State and sheep in FCT. Age-specific prevalence of cysts in the Sheep showed highest prevalence in 3-4 years age group (12.8%) of sheep and 1-2 years age group (11.7%) of Goats, difference in prevalence of cysts among different age groups of Sheep and Goats were not significant ($p>0.05$). Prevalence of hydatid cysts was significantly associated with 3-4 years age group of Sheep (OR=4.015, 95% CI=2.100-7.675) and 1-2 years age group of Goats (OR=1.923, 95% CI=1.392-2.658). More hydatid cysts were recovered in female sheep (10.7%) and male Goats (11.6%) than their counterparts, the difference was not significant ($p>0.05$) in Sheep but in Goats ($p<0.05$). Female Sheep and male Goats were more likely to harbour hydatid cysts (OR=1.752, 95% CI=0.984-3.121; OR=1.884, 95%CI=1.363-2.604). Higher numbers of fertile cysts were recovered from liver and lungs of Sheep (34.3%) than Goats (33.3%). Fertility of hydatid cysts is in the epidemiology and control of hydatidosis and results of the present study has provided a base line data on hydatidosis in sheep, goats, and cattle slaughtered in selected abattoirs in northern Nigeria which will help in controlling the disease.
Keywords: Prevalence, Hydatid cysts, Sheep, Goats, Northern Nigeria.

1.0 Introduction

Hydatidosis also called Cystic Echinococcosis is a major parasitic zoonotic disease of veterinary and public health importance caused by the larval stage of *Echinococcus granulosus sensu lato* (Soulsby, 1982). The disease has a worldwide distribution and prevalence of infection differs from one region to the other with higher prevalence in populations that practice animal husbandry (Dada & Belino, 1978). The disease is endemic in some parts of Asia, Middle East, South America and Africa (Regassa, 2019; Ohiolei *et al*.,

2020) and is reported more commonly in food animals that are communally owned or raised on free range and associated more intimately with domestic dogs due to poor handling practices (Grosso *et al*., 2012). Dogs and carnivores are the definitive hosts of *Echinococcus granulosus* while a wide range of herbivores including domestic animals (Sheep, Goat, Pigs, Cattle, Camels, Horses) act as the intermediate hosts; humans are accidental intermediate hosts (Eckert and Deplazes, 2004; Founta *et al*., 2016).

Livestock acquire the infection by ingesting infective eggs in forage which later develop into larval stages in different organs of their bodies (Soulsby, 1986; Thompson, 1986). The life cycle is completed when fertile hydatid cysts are ingested by the definitive hosts and develop to adult worms. Humans get infected during natural transmission of the disease from carnivores to domestic animals by ingestion of eggs of *Echinococcus* in contaminated food, water or through direct contact with dogs (Abriham, 2021).

The disease is characterized by formation of variable hydatid cysts in visceral organs of the intermediate hosts and adult tapeworm in the intestine of dogs (Thompson & McManus, 2001). Hydatid cyst is the larval metacestode that develop as the infective stage in internal organs of the intermediate hosts, most commonly in the liver (50-70%) and less commonly in lungs, spleen, kidney and brain (Botezatu *et al*., 2018). The disease has an adverse effect on livestock production thereby causing decreased production of meat, milk, wool and reduction in growth rate alongside predisposition to other diseases (Soulsby, 1982).

In Nigeria, livestock rearing besides meeting human needs is occupational especially in Northern Nigeria and parts of the south where pastoralists migrate in search of food and water for their herds (Ohiolei, 2020). over time, livestock population have risen significantly with estimates of 19 million cattle, 37million sheep, 65 million goats, 6 million pigs, 1 million donkeys and 277,000 camels (National Agriculture Sample Survey, Nigeria, 2010). Despite the increase in livestock population in Nigeria, the economic impact of hydatidosis on livestock production is not evaluated as uninvestigated in most states (Ohiolei *et al*., 2019). Reports during the last three decades in few areashave shown high prevalence in livestock raised in Northern Nigeria (Dada and Belino, 1978; Dada *et al*., 1980; Ogunsan *et al*., 2000; Tijjani *et al*., 2010; Bello *et al*., 2020; Okolugbo *et al*., 2023). It is with this background that the present study was embarked upon to determine the current prevalence of hydatidosis and the fertility rate of hydatid cysts in sheep and goats slaughtered at selected abattoirs in Northern Nigeria.

2.0 Methods

2.1 Study area and population

The study was carried out in selected abattoirs located in four States (Gombe, Kogi, Plateau and Kaduna) in Northern Nigeria and the Federal Capital Territory, Abuja, Nigeria (Fig. 1). The study population consisted of Sheep and Goats brought for slaughter. The liver and lungs from infected animals were examined.

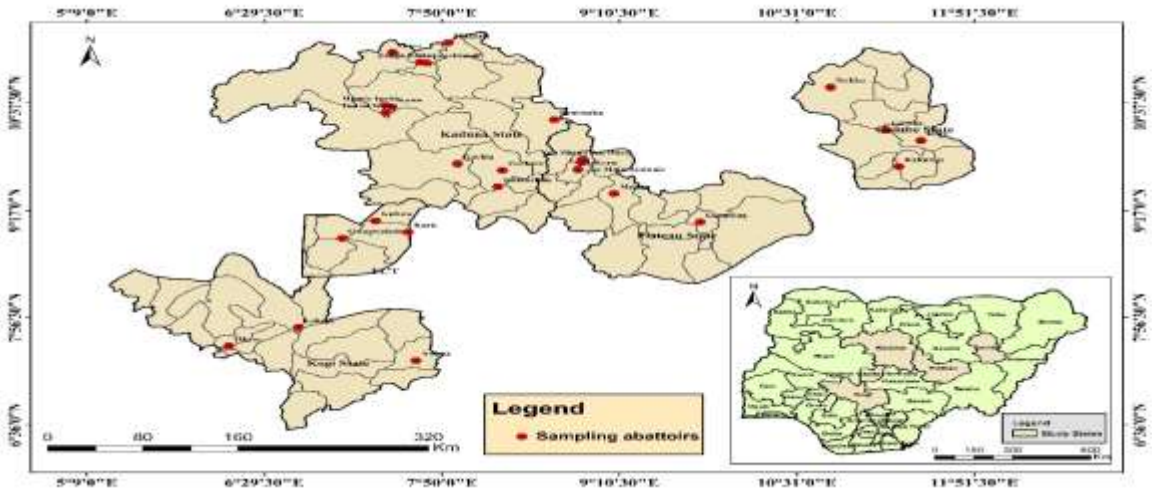


Figure 1: Map of the Study Area

2.2 Sample Collection

Purposive sampling method was used to collect samples from sheep and goats slaughtered in selected abattoirs. Prior to collection of samples, information were obtained on the animals and abattoirs which included sex, estimated age of the animals, presence or absence of stray dogs around the abattoir, general hygiene of the abattoir and information on whether the abattoir is fenced or not.

2.3 Ante Mortem Examination

Ante mortem examination was performed before slaughter and the ages of the animals were estimated on the basis of the dentition by eruption of pinchers, 1st intermediate pair, 2nd intermediate pair of large permanent incisors which indicates 1 year, 2years, 3years up to 5 years and above Eruption and eventual loss of teeth indicated 5years and above and were grouped as follows; 1 to 2 years, 3 to 4 years, and 5 years and above (Delahunta and Habel, 1986).

2.4 Post Mortem Examination

According to the technique described by Fathi *et al*. (2011), post-mortem examination of the animals brought for slaughter at the abattoir was carried out through visual inspection and palpation of the lungs and liver of the study population. Each animal was carefully examined for hydatid cysts which if found were removed from the infected organs, preserved in clean sample bottles with labels and sent to the Parasitology and Entomology Research Laboratory, Department of Zoology, Ahmadu Bello University, Zaria for further analysis.

2.5 Laboratory Analysis (Cyst characterization)

Individual cysts recovered from each animal were carefully incised and examined microscopically. Cysts were categorized as fertile if they had protoscoleces which appeared as white spots on the germinal epithelium in hydatid fluid while cysts that were gritty on incision were classified as calcified (Soulsby, 1982).

2.6 Data Analyses

Data obtained were analyzed using the Epi-Info software version 7.0 for epidemiological studies. Significance of difference in prevalence in relation to age, sex, predilection sites (liver and lung) in the infected sheep and goats were determined using Chi-square (χ^2) analysis while the significance of associations of prevalence with the stated risk factor variables were determined using Odds Ratio (OR) analysis. Association of prevalence of infection with a risk factor was present if the OR value was greater than one (>1.0) and was absent if the OR

value was less than one (<1.0). Associations were significant when the lower limit of the 95% confidence interval of the OR value was >1.0 and was not significant if it was <1.0.

3.0 Results

3.1 Overall prevalence of hydatid cysts in Sheep and Goats

An overall prevalence of hydatid cysts in liver and lungs of 2,830 livestock comprising 906 sheep and 1,924 goats was 14.3%. Hydatid cysts were detected in 87 (9.6%) sheep and 175 (9.1%) goats, the difference was not statistically significant (p>0.05) (Table 1).

3.2 Prevalence of hydatid cysts in Sheep and Goats by location

The prevalence of cysts based on location revealed higher prevalence in 38.9% of sheep and 16.0% of goats slaughtered in selected abattoirs in Kaduna State followed by 23.0% in sheep and 13.0% in goats slaughtered in Gombe State, least prevalence's of 3.0% each was obtained in Goats slaughtered in Plateau State and Sheep in FCT. The difference of prevalence of Sheep and Goats among the States was statistically significant (p<0.05). There was significant association between prevalence of hydatidosis in Sheep and Goats with location in Gombe (Sheep-OR=7.564, 95%CI=4.608-12.416; Goat- OR=1.628, 95% CI=1.114-2.379) and Kaduna State (Sheep-OR=6.948, 95%CI=3.410-14.155; Goats-OR=4.462, 95% CI=3.144-6.333). There was no association between hydatid cyst infection with location in Sheep slaughtered in Plateau and FCT and goats slaughtered in Plateau State whose OR values were <1.0 (Table 2).

Table 1. Overall prevalence of hydatid cysts in sheep and goats slaughtered in selected abattoirs in Northern Nigeria

Species	Number Examined	Number Infected (%)	Chi square	P. Value	OR (95% C.I.)
Sheep	906	87 (9.6)	0.188	0.664ns	1.062 (0.810 – 1.391)nsa
Goats	1924	175 (9.1)			0.942 (0.719 – 1.234)nsa
Total	**2830**	**262 (14.3)**			

Key: ns=not significant difference, nsa =not significant association, OR= Odds Ratio

Table 2. Prevalence of Hydatid cysts in cattle slaughtered in selected abattoirs in Northern Nigeria

States	Number Examined	Number Positive (%)	Chi-square	p Value	OR (95% C.I.)
Goat					
Kogi	309	0 (0.0)	121.452	<0.001*	NA
Gombe	301	39 (13.0)			1.628 (1.114 - 2.379)sa
FCT	275	0 (0.0)			NA
Plateau	235	7 (3.0)			0.278 (0.129 - 0.600)na
Kaduna	804	129 (16.0)			4.462 (3.144 - 6.333)sa
	1924	**175 (9.1)**			
Sheep					
Kogi	268	0 (0.0)	136.865	<0.001*	NA
Gombe	274	63 (23.0)			7.564 (4.608 - 12.416)sa
FCT	266	8 (3.0)			0.220 (0.105 - 0.462)na
Plateau	62	2 (3.2)			0.298 (0.072 - 1.240)na
Kaduna	36	14 (38.9)			6.948 (3.410 - 14.155)sa
	906	**87 (9.6)**			
Total	**2830**	**262(14.3)**	**0.188**	**0.664ns**	

3.3 Age-specific prevalence of hydatid cysts in Sheep and Goats

The Overall age-specific prevalence of cysts revealed higher prevalence in Sheep (9.6%) than Goats (9.1%). Highest prevalence of 12.8% was recorded among Sheep of 3-4 years age group and 11.7% among goats of 1-2 years age group while the least prevalence was 3.4 % in 1-2 years age group of Sheep and 5.3% in Goats of 5 years and above age group. The difference in prevalence of hydatid cysts among different age groups of sheep and goats were statistically significant ($p<0.05$). Prevalence was significantly associated with Sheep in 3-4 years age group (OR=4.015, 95%CI= 2.100-7.675) and Goats in 1-2 years age group (OR=1.923, 95% CI=1.392-2.658) (Table 3).

3.4 Sex-specific prevalence of hydatid cysts in Sheep and Goats

Sex-specific prevalence of hydatid cysts was higher in female sheep (10.7%) than males (6.4%), the difference was not statistically significant ($p>0.05$) while in Goats, prevalence was higher in males (11.6%) than females (6.5%) and the difference was statistically significant ($p<0.05$). Female sheep (OR=1.752, 95% CI=0.984-3.121) and male goats (OR=1.884, 95% CI=1.363-2.604) were more likely to harbour the cysts than their counterparts (Table 4).

Table 3. Age-specific prevalence of hydatid cysts in sheep and goats slaughtered in selected abattoirs in Northern Nigeria

Species/ Age Groups (Years)	Number Examined	Number Positive (%)	Chi-square	P. Value	OR (95% C.I.)
Sheep					
1 to 2	290	10 (3.4)	20.274	<0.001*	0.250 (0.127 – 0.491)na
3 to 4	594	76 (12.8)			4.015 (2.100 – 7.675)sa
5 and above	22	1 (4.5)			0.442 (0.059 – 3.326)na
	906	**87 (9.6)**			
Goat					
1 to 2	964	113 (11.7)	17.240	<0.001*	1.923 (1.392 – 2.658)sa
3 to 4	547	40 (7.3)			0.726 (0.503 – 1.048)nsa
5 and above	413	22 (5.3)			0.499 (0.315 – 0.792)na
	1924	**175 (9.1)**			

Key: ns=not significant difference, *=significant difference, sa=significant association, nsa=not significant association, na=no association, NA=not applicable

Table 4. Sex-specific prevalence of hydatid cysts in sheep and goats slaughtered in selected abattoirs in Northern Nigeria

Species/Sex	Number Examined	Number Positive (%)	Chi-square	p Value	OR (95% C.I.)
Sheep					
Male	234	15 (6.4)	3.704	0.054ns	0.571 (0.320 – 1.017)na
Female	672	72 (10.7)			1.752 (0.984 – 3.121)nsa
	906	**87 (9.6)**			
Goats					
Male	973	113(11.6)	10.094	<0.001*	1.884 (1.363 – 2.604)sa
Female	951	62(6.5)			0.531 (0.384 – 0.734)na
	1924	**175 (9.1)**			

Key: ns=not significant difference, *=significant difference, sa=significant association, nsa=not significant association, na=no association, OR=Odds Ratio

3.5 Prevalence of cysts in liver and lungs of Sheep and Goats

Out of 2,830 Sheep and Goats examined, 262 representing 9.3% of the study population harboured cysts in liver (6.7%), lung (3.6%) or both (1.0%), with highest prevalence in sheep (9.6%) than goats (9.1%) but difference in prevalence of cysts in Goats and Sheep was not statistically significant (p>0.05). Highest numbers of cysts were recovered in liver of Sheep (7.3%) and Goats (6.5%) than lungs of both animals (3.6% in both) and only 1.0% of the infected sheep and goats had cysts in liver and lungs (Table 5).

Table 5. Overall prevalence of cysts in liver and lungs of sheep and Goats slaughtered in selected abattoirs in Northern Nigeria

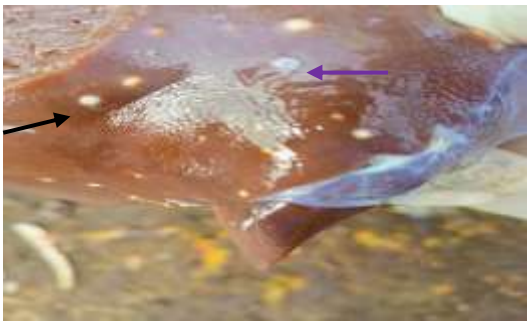
Animals	Number Examined	Prevalence (%)			
		Overall	Liver	Lungs	Lungs and Liver
Goats	1924	175 (9.1)	125 (6.5)	70 (3.6)	17 (0.9)
Sheep	906	87 (9.6)	66 (7.3)	33 (3.6)	10 (1.1)
Total	**2830**	**262 (9.3)**	**191 (6.7)**	**103 (3.6)**	**27 (1.0)**
Chi-square		0.188	0.608	0.000	0.316
Df		1	1	1	1
p Value		0.664ns	0.436ns	0.996ns	0.574ns

3.6 Characterization of hydatid cysts obtained from liver and lungs of Sheep and Goats

In relation to cyst fertility, a total of 99 (10.9%) sheep and 195(10.1%) Goats harbored cysts in their liver and lungs; fertile cysts were recovered in 34.3% of infected sheep and 33.3% infected goats. The difference in prevalence of hydatid cysts in liver and lungs of sheep was not significant (p>0.05) but significant in goats (p<0.05). More sheep and goats harboured fertile cysts in their liver (39.4%, 41.6%) than their lungs (24.2%, 18.6%). However, the rate of calcification was higher in lungs of Goats (81.4%) than Sheep (75.8%) (Table 6). The characterization of hydatid cysts in liver and lungs is illustrated in Figure 2.

Table 6. Viability of Cysts in liver and lungs of sheep and goats slaughtered in selected abattoirs in Northern Nigeria

Animal	Number with Cysts (%)	Number with Fertile Cyst (%)	Number with Calcified Cyst (%)	Chi-square	p Value
Sheep (n = 906)					
Liver	66 (7.2)	26 (39.4)	40 (60.6)	2.240	0.134ns
Lungs	33 (3.6)	8 (24.2)	25 (75.8)		
Total	**99 (10.9)**	**34 (34.3)**	**65 (65.7)**		
Goats (n = 1924)					
Liver	125 (6.5)	52 (41.6)	73 (58.4)	10.708	0.001*
Lungs	70 (3.6)	13 (18.6)	57 (81.4)		
Total	**195 (10.1)**	**65 (33.3)**	**130 (66.7)**		



(a)

(b)

(c)

Figure 2. Hydatid cysts in liver and lungs of slaughtered ruminants; (a) Liver of Goat showing (i) fertile cyst ← and (ii) sterile cyst ↗ (b) lung of sheep showing fertile and calcified cysts (c) liver of Sheep showing multiple cysts (calcified and fertile)

4.0 Discussion

Hydatidosis is a severe health problem that is prevalent in many parts of the world including Nigeria. The study indicated that sheep (9.6%) had a greater prevalence of hydatidosis than goats (9.1%), albeit the difference was not statistically significant. The prevalence was lower than 11.4% in sheep and 55.5% in goats slaughtered in Kano Luka *et al*. (2010), 19.6% in sheep and 16.0% in goats slaughtered in Addis Ababa abattoir by Kebede *et al*. (2010) prevalence of 72.2% and 58.4% was obtained in sheep and goats slaughtered in Southern Italy by Bosco *et al*. (2021). Higher than the 4.25% in sheep and 0.64% in goats obtained by Meerkhan *et al*. (2018) in Iraq, 0.07% in sheep and 0.01% in goats obtained by Tijjani *et al*. (2010) in Damaturu, Yobe State, Nigeria, and 1.71% in sheep and 2.14% in goats obtained by Bello *et al*. (2020) in Adamawa State, Nigeria. The highest prevalence of hydatidosis among sheep in this study suggests that sheep are highly susceptible to the parasite.

Generally speaking, differences in the prevalence of hydatidosis reported in various localities depend on the availability of the host, resource-poor setting, pastoral lifestyle, changes in the environment that favor egg survival, transboundary animal movement and livestock trade, presence of stray dogs, dog-man contact, and poor condition of the abattoir in the area (Anyanwale *et al*., 1982; Ohiolei *et al*., 2020). To our knowledge, this survey is the first to document the prevalence of hydatidosis in sheep and goats in Gombe, Kaduna, Kogi and Plateau States and FCT in Northern Nigeria. The highest prevalence of hydatid cysts has been found in Gombe State, which may be related to the state's largely free-range animal husbandry system, low public awareness of the disease and indiscriminate and illegal animal slaughter brought on by a lack of modern abattoirs in most of the study locations. Similar findings have also been reported by other researchers (Okolugbo *et al*., 2023; Tijjani *et al*., 2010; Dada and Belino, 1978).

Most rural communities surveyed for this study in all the states have just one slaughterhouse that cannot handle hundreds of animals killed every day, forcing some residents to slaughter their animals at home. Infected viscera that are a major factor in the maintenance and transmission of the cyclozoonotic disease in domestic ruminants and humans are exposed to stray dogs due to the complete lack of meat inspection at slaughter and backyard slaughter practices (Regassa, 2019). The fact that the Federal Capital Territory's (FCT) abattoirs are well-equipped, cleaner, and properly fenced, in addition to the routine meat inspections conducted by veterinary doctors at the abattoirs, may be the cause of the lowest prevalence recorded in sheep and zero prevalence in goats slaughtered in the FCT. As a result, in the abattoirs, only animals that appear healthy are sent to be slaughthered.

Age groups of 1-2 year old goats and 3-4 year old sheep showed higher prevalence. This result may be attributable to the fact that animals of various age groups were exposed to comparable animal husbandry techniques with comparable environmental conditions, giving them an equal chance of coming into contact with the infective stage of *Echinococcus* species in their communities. Male goats had more cysts than females, while female sheep had more cysts than male sheep in every slaughterhouse. This implies that exposure to the same risk factors for both sexes suggest that sex may not be a predisposing factor in the development of the disease Tijjani *et al*. (2010).

In this study, hydatid cysts were recovered more frequently from sheep and goat livers than from lungs, which is consistent with other findings from around the world (Saulawa *et al*., 2011; Muqbil *et al*., 2012; Regassa, 2019; Abriham, 2021; Okolugbo *et al*., 2023) The most frequent predilection sites in domestic animals are the liver and the lungs. This is because these organs have many capillaries, which facilitate the efficient filtration of ingested onchosphere from the blood and the subsequent displacement of cells in the organs by formation of a fibrous capsule around the parasite, which enlarges to accommodate cyst as it grows. Studies have revealed that the cyst may occasionally localize in the kidney, spleen, or brain tissues, where its effects are more severe and fatal (DNR, 2009; Abriham, 2021)

As all blood from the gastrointestinal tract passes through the liver before reaching the lungs, more onchospheres are trapped in the liver than the lungs, which is why more fertile cysts were found in the livers of sheep and goats. The primary organ for detoxification being the liver, cysts there trigger an immune response that causes the cysts to calcify. (Baldock *et al*., 1985; Njoroge *et al*., 2002; Kebede *et al*., 2009; Muqbil *et al*., 2012). The liver contained more calcified cysts, which may have been caused by the liver's abundant connective tissue reaction and relatively higher reticuloendothelial cell density (Arene, 1985; Thompson, 1986; Getachew and Angesom, 2019).

Different animal origins, strains of *Echinococcus granulosus*, grazing techniques and other environmental factors may have contributed to the study's variability in the fertility of cysts collected from various locations (Gachengo *et al*., 2017). Muqbil *et al*. (2012) speculated that differences in tissue resistance in the various organs could account for the viability in fertility of cysts isolated from liver and lungs of sheep and goats and this is in line with the observation of Getachew & Angesom (2019) and Ohiolei (2019).

5.0 Conclusion

This study have revealed that hydatidosis is highly prevalent zoonotic disease of unrecognized importance in Nigeria that is causing serious economic losses due to organ condemnation in the abattoirs. This is due to the presence of stray dogs (definitive host) around the unfenced abattoirs which play a significant role in transmission of the disease. Moreover, lack of public health awareness about the disease, lack of infrastructures and services for proper meat inspection at all levels, traditional animal husbandry and backyard slaughtering of animals have contributed to a great extent to occurrence of the disease. Our findings also confirm that fertile cysts recovered from liver and lungs of sheep and goats indicates that these ruminants are among the major intermediate hosts responsible for perpetuation and maintenance of life cycle of hydatidosis in Nigeria especially in communities where there is close contact between the

definitive and intermediate hosts. Information obtained from this study could serve as baseline data for further studies thereby enriching the data on impact of hydatidosis in Nigeria. We suggest that areas that have not been investigated should be studied in order to provide enough data for control of this zoonosis of unrecognized importance in Nigeria.

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Production of Auto Connecting Rod from Scrapped Steel and Palm Kernel Shell

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Abstract
Connecting rod significantly impacts both piston and crankshaft, it acts as an intermediate link resisting tensile and compressive forces. Palm Kernel Shells (PKS) are solid, firm, carbonaceous, and organic by-products of the processing of the palm oil nut. This research aimed to produce an auto connecting bar from scrapped steel and PKS. The PKS were dried and milled into powder, charged into a rotary furnace containing molten scrapped steel. A connecting rod pattern is made using a sand mold after modelling using Auto desk inventor. The hot liquid ingot is poured to fill up the mold space. The cast is removed after cooling and prepared for Impact, Tensile and Hardness examinations. Samples of the product are also tested using Scanning Electron Microscope (SEM) furnished with Energy Dispersive Spectroscopy (EDS). The results showed that the maximum impact strength is at interface friction of about 32.85o, Internal friction angle of 19o and a tangential force of about 0.35 kN and yields at a tensile stress of 272.0 MPa. The brinel hardness value of 245 MPa was obtained. The SEM revealed graphite precipitates in mixture of pearlite and ferrite. The EDS Analysis showed the presence of 4.30% Carbon, 1.3 % Silicon, 0.54% Copper, 0.25% Nickel, 6.20% Magnesium, 85% Iron, 0.15% Manganese, 1.22% Zinc and 0.30% Chromium. This study confirmed that the connecting rod produced can withstand the necessary tensile and compressive loads.
Keywords: Cast iron, Connecting rod, Palm Kernel Shell, Characterization, Mechanical Property

1.0 Introduction

Connecting rod significantly impacts both piston and crankshaft, it acts as an intermediate link resisting tensile and compressive loads. A tension load connecting rod is made of either forged steel, cast steel, or fabricated steel while rods with a compression loading are cast nodular steel or aluminum alloy. Connecting rod can be either H-Beam or I-Beam type. H-Beam can handle more stress without bending and is used in high power engines. I-Beam is both light weight and strong but the type of material used limits its capacity to handle loads **(**Siddique and Reddy, 2015**).** Casting involves heating the metal alloy involved until it is molten, and pouring the molten metal alloy into "sprues" which are joined to "runners" channeling the molten metal alloy into a multiple cavity mould. The molten metal alloy fills the mould, and vents through "risers" the metal casting is separated from the mould upon cooling, the sprues, runners, and risers are removed, and the casting is machined for good surface finish **(**Akande *et al*., 2023**).**

Rao *et al.*,2013 studied the weight reduction opportunities in the connecting rod of an Internal Combustion engine by examining various materials such as Genetic Steel, Aluminium, Titanium and Cast Iron. This was entailed by performing a detailed load analysis comprising static load and stress analysis of the connecting rod, and design optimization for suitable material to minimize the deflection. Verma and Jain, 2022 described the modelling and analysis of connecting rod. In the research, connecting rod was replaced by Aluminium reinforced with Boron carbide for Suzuki GS150R motorcycle. A connecting rod

was designed (Nitturkar *et al.*, 2020) using aluminium alloy instead of steel and Finite Element Analysis approach was explored in the analysis to determine the various stresses. It was observed that in many cases weight reduction of connecting rod was obtained by replacing a material with lighter one. The widely used materials in connecting rod manufacturing are carbon steel, cast iron, wrought steel or powder metal etc. The scope thus exists to try other materials like Titanium alloy, carbon fiber, aluminium alloy, glass fiber etc. to produce light weight alternative because they are light in weight. Therefore, optimizing the connecting rod for weight reduction with the use of such materials can be explored. The connecting rod is a determinant factor in the efficiency of the engine. Other factors which affect the efficiency and performance of the engine included the material of the connecting rod, its weight and the amount of power used up to move the connecting rod in the power stroke (Sriharsha *et al.*, 2021).

The Oil Palm is an important tree in Nigeria and the PKS as residue has been regarded as 'waste' from palm oil processing (Adetunji *et al*.,2022). The palm oil industry produces wastes such as palm kernel shells and palm oil fibres, which are usually dumped in the open thereby impacting the environment negatively without any economic benefits. The PKS are solid, firm, carbonaceous, and organic by-products of the processing of the palm oil fruit. The PKS consists of small, medium and large size particles in the range 0.5, 5-10 and 10-15 mm **(**Haider *et al.*, 2018**)**.

Reduction of waste palm kernel shell and unused scrapped auto engine will make the environment neater coupled with promoting small scale industry to build Nigerian economy. This study produced a connecting rod from palm kernel shell (PKS) as carbon source and scrapped steel using sand casting with rotary furnace.

2.0 Materials and Methodology

This research used PKS, Scrapped Steel and moulding Sand.

2.1 Designing the connecting rod using Autodesk inventor

The auto connecting rod was designed using the software package.

2.2 Casting of the auto connecting rod

The powdered PKS is prepared by drying, heating and grinding. The sand mould surface was smoothed to accommodate the pattern. A sample of the connecting rod is pressed tightly on the soft ground to make a pattern on the sand, which is the method used to make the part. The top part of the mould is a wooden structure which has been pre-filled with a special sand that stays permanently on the wooden frame, the pattern is formed on the top part when the sample is placed carefully on the base of the mould and also pressed tightly, so that the pattern is formed. On the top part of the mould some holes are intentionally made in order to pour the molten metal through and allow it to fill up the pattern.

The scrapped steel is charged in a rotary furnace and the temperature was set at 1700^{o}C and maintained for six hours, then the prepared PKS powder was charged to the melt and stirred rigorously to make it mix evenly with the molten metal, after this was done, then the metal ingot was poured into the sand mould with the rod pattern, and allowed to cool for about 24 hours after which the casting was taken out.

2.3 Investigation of mechanical property

The impact stress, tensile stress and hardness tests of the produced auto connecting rod samples were carried out on Triaxial Impact Machine, Universal Testing Machine and Brinell hardness tester, respectively. The Brinell Hardness number is obtained from BHN=F/πDh (Brines Hardness Number)

2.4 Morphological characterisation

The structure and phase evolution of the sample produced was characterised using scanning electron microscope.

3.0 Results

3.1 Orthographic view of the connecting rod

Fig. 1 contains the orthographic view of the auto connecting rod produced.

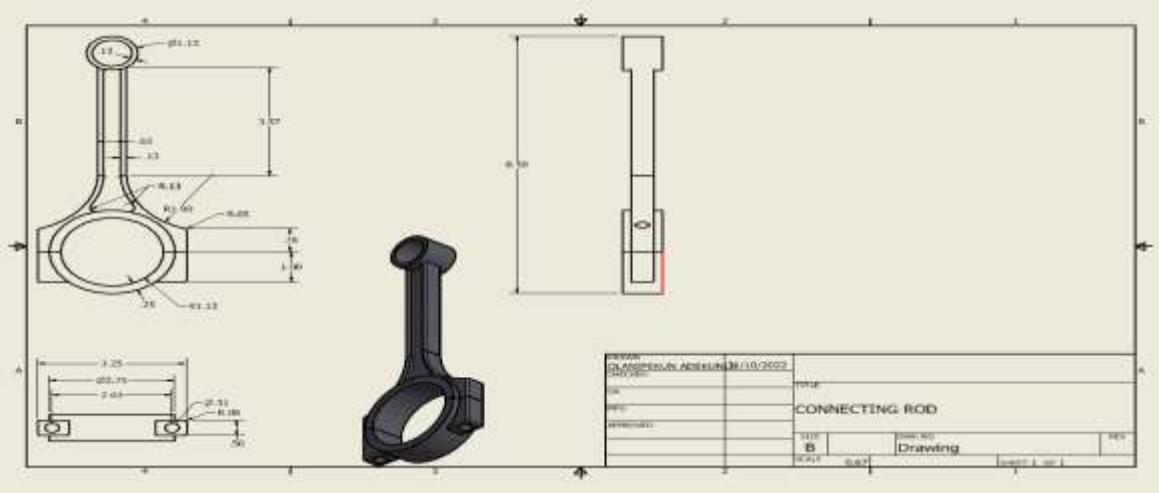

Fig.1 : Orthographic views of the auto connecting rod

3.2 Cast result of the connecting rod

Figure 2 contained the cast connecting rod produced.

Fig. 2 Connecting rod cast sample

3.3. Impact Test Result.

A maximum impact strength of 130.4 kN/m^2 was obtained at penetration of 2.5 mm under impact force of 160 kN/m^2. Other results are contained in Table 1. The impact test is obtained using Triaxial Impact Machine, the results are shown in the table below .

Table 1 Impact result of connecting rod at different loads

6_3 = 80 kN/m^2				6_3 = 160 kN/m^2				6_3 = 320 kN/m^2			
Penet. (mm)	Load N	Strain X 10^{-2}	Stress KN/m^2	Penet. (mm)	Load N	Strain X 10^{-2}	Stress KN/m^2	Penet. (mm)	Load N	Strain X 10^{-2}	Stress KN/m^2
0	0	0	0	0	0	0	0	0	0	0	0
0.40	4	0.40	3.52	0.30	10	0.44	8.77	0.22	5	0.30	4.40
0.55	30	0.74	24.53	0.48	20	0.70	17.50	0.42	10	0.60	8.80
0.66	56	0.92	41.87	1.00	92	1.42	81.72	0.94	10	1.30	10.44
1.20	78	1.40	62.52	1.54	119	2.12	103.40	1.10	14	1.50	13.90
1.58	120	2.13	93.23	2.04	137	2.65	115.62	2.20	30	1.05	24.20
1.94	140	2.73	115.24	2.50	150	3.52	130.40	2.88	60	3.90	47.50

3.3. Tensile Strength Result

Table 2 contained the tensile strength (i.e Frictional Angle and deformation modulus strength of the sample) of the sample.

Table 2:Tensile strength results

Stress (MPa)	Normal Force (KN)	Tangential Force (KN)	Either Force	Friction Angle (°)Int.face	Intern. Frict' Angle (°)	Tensile Strength (MPa)	Deformation Modulus (MPa)
0	0.1	0.1	1.00	22.20	19	0	0
20	0.40	0.22	1.75	34.60	19	170. 45	210.43
35	0.45	0.35	1.28	32.85	19	272.60	254.54
45	0.27	0.24	1.00	24.90	19	230.54	340.20

3.4 Brinel Hardness Test Result

The resistance of the sample to indentation and abrasion is calculated by brinel hardness test, Table 3 contained the result.

Table 3: Hardness test result

Indenter	Force (KN)	Diameter of ball(mm)	Diameter of indentation(mm)	Depth of indentation(mm)	Brinel Hardness Number
Tungsten Carbide	7.7	10	3	1	245

The Brinel Hardness Number is **245.07.**

3.5 Scanning Electron Microscope(SEM) Results

The scanning electron microstructure of the cast sample is shown in Fig.3, the SEM images shown below. The SEM revealed graphite precipitates in mixture of pearlite and ferrite.

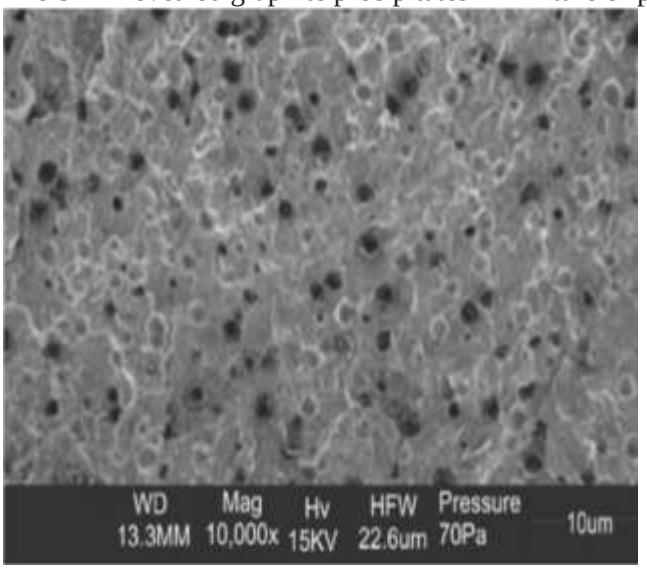

Fig. 3: SEM result of the sample

The diagram(Fig.4) shows the Energy Disperssive Spectroscopy (EDS) Analysis of the connecting rod, showing that its carbon content is about 4.30%, confirming the microstructure of cast iron.

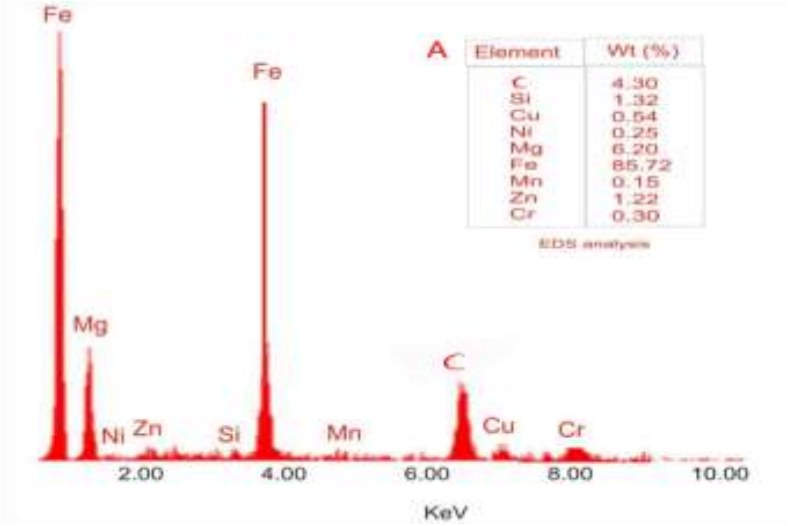

Fig. 4: EDS of the connecting rod sample.

4.0 Discussion of Results

The quantitative result of the impact tests the energy needed to fracture a material and can be used to measure the toughness of the material. There is a connection to the yield strength but it cannot be expressed by a standard formula**.**The result of the impact test shown in Table 1, shows the stress at different impact Pressures, 80 kN/m^2, 160 kN/m^2 and 320 kN/m^2 respectively. The results show that the maximum impact strength of 160 kN/m^2 of 2.5 mm penetration at interface friction of about 32.85^o, Internal friction angle of 19^o and a tangential force of about 0.35 kN and yields at a tensile stress of 272.0 MPa.

The maximum tensile strength of the component according to the result is 272.60 MPa, this value is as a result of a high carbon content of the sample (Kuye *et al.*, 2021). The brinel hardness number for ductile cast iron is about 235 BHN, as reported in some literature while 245 BHN was gotten in the investigation confirming the cast iron product.

The EDS Analysis result showed that the connecting rod consisted of 4.30% Carbon, 1.3 % Silicon, 0.54% Copper, 0.25% Nickel, 6.20% Magnesium, 85% Iron, 0.15% Manganese, 1.22% Zinc and 0.30% Chromium. The most important constituents in this mixture is the Percentage of Iron, Silicon and Carbon, which are commensurate to that of cast Iron, this result shows that this material is a cast Iron.

The SEM results shown in Fig. 3 , showed that the sample exhibits a fine surface texture, this result is similar to that of (Akande, 2023), it also indicates the presence of grain refining elements such as C, Si, Mg, and Zn. In the EDS analysis the sample as shown in Fig.4, it was observed to contain a high percentage weight of Carbon, indicating possibility of better mechanical properties such as, hardness, and strength making it more resistant to wear and abrasion.

It is observed that there are some dark round spots on the image, which depict graphites in transformed austenite and cementite matrix. The graphite flakes have low density hence compensate for the freezing

contraction thus enhance the quality of the casting (Adetunji *et al*., 2022). The EDS analysis carried out by Akande *et al*., 2023, is quite similar to this, but differed because of the goat bone used as a reinforcer of the cast iron thereby increasing its calcium and zinc content, asides this the result from this EDS is similar in terms of carbon composition.

5.0 Conclusion

An auto connecting rod was produced from scrapped steel and Palm kernel shell through casting in a rotary furnace. The followings were concluded.

1. The auto connecting rod produced from the scrapped steel and Palm kernel shell is suitable for the replacement of the part in motor cycle.
2. The impact, tensile and hardness properties are adequate for the motor cycle requirement with maximum values of 160 kN/m^2, 245 BHN and 272.6 MPa, respectively..
3. The scanning electron image obtained and the energy disperssive spectroscopy results revealed graphite precipitates in mixture of pearlite and ferrite, and 4.3% carbon content.

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Cost Analysis of Heat Detection and Performance of Sows using Teaser Boars and BOARBETTER® Pheromone Post-Weaning

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Abstract
A study was carried out to investigate the cost analysis of heat detection and performance of sows using teaser boars (TB) and BOARBETTER® (BB) pheromone post-weaning at Livingston's Enterprises Inc., Fairbury, Nebraska, USA. A total of 4,810 F_1 (American yorkshire [sow] X landrace [boar]) weaned sows were used. The sows were allotted into two treatment groups (group 1 (control): sows heat-checked using TB exposure and group 2: sows heat-checked using BB exposure) in a student t-test. Feed and water were given appropriately. Data were collected on sow performance and piglet performance, while cost effectiveness was calculated. Data were analyzed using Statistical Package for Social Sciences, version 20. Significant ($P<0.05$) differences occurred in weight at birth (WB), weight at weaning (WW), piglets weaned per sow (PWS), weight gain (WG) and feed conversion ratio (FCR). WB was higher in control (0.68 kg), WW was higher in BB (4.54 kg), PWS was higher in BB (15.58), WG was higher in BB (3.63 kg) and FCR was lower in BB (3.50). Pheromone was twice more efficient than TB in all the parameters considered. It was concluded that both from production and economic point of view, BB was more efficient than TB in pig reproduction.
Keywords: swine reproduction, teaser boars, BOARBETTER®, heat detection

Introduction

Although pigs are quite abundant, poor management can reduce their ability to reproduce. (Njoga et al., 2021). Pigs are mostly kept for their meat and the most important trait of interest for their genetic improvement is the body weight. Body weight is an economically important trait (Cam et al., 2010). Pigs are raised for festivals (Ate and Oyedipe, 2011), home consumption and for financial security (Nwanta et al., 2011). Pigs are omnivores that are raised primarily for their meat, manure and as a source of biomedical raw materials (insulin) across most of the world (Ajibo et al., 2020).

Thankfully, because of their polytocous nature, pigs have the potential to become the most popular animal protein-deficient gap filler in Nigeria (Pierozan et al., 2016). Furthermore, pig farming can be a substantial source of labor for the livestock subsector, which can help developing nations reduce crime, create jobs, and reduce poverty (Pierozan et al., 2016).

Pigs have comparative advantages over other meat-producing animals due to their high feed conversion efficiency, early maturing nature, short gestation length (Njoga et al., 2021), and capacity to thrive in less-than-ideal tropical conditions (Pierozan et al., 2016). These attributes also support the potentials mentioned earlier. Moreover, pig farming offers a spatially efficient alternative to the production of ruminants. Furthermore, the pig industry's marketability of pork and pork products tends to ensure timely returns on investment and profitability (Nwanta et al., 2011). From an economic perspective, the most significant

indicator of productivity in breeding herds is the number of piglets weaned per sow year (Ate and Oyedipe, 2011).

A teaser boar is a male pig rendered sterile following vasectomy or epididymectomy, but still hormonally and behaviourally sexually active. Teaser boars are employed to spread pheromones near gilts and sows. The use of live adult males delivers variable concentrations of pheromones, provide visual, auditory and at times tactile signals to the sows. The cost of keeping and sustaining such population of teaser boars and breeding boars is usually overwhelming.

The role of boars in pig herds was discussed by Langendijk et al. (2002). Farmers have been known to be deterred from raising breeding boars in the past by a variety of issues, such as the animals' propensity for aggression, the expense of feed, and the time required to care for the boar. Perhaps the primary cause of the decline in household boar ownership is the high expense of raising boars (Mutua et al., 2011).

In the study by Wabacha et al. (2004), In Central Kenya, farmers view raising boars as unprofitable and instead choose to employ hired boars rather than breeding their own. Due to the associated expenses, farmers in Nepal are hesitant to continue breeding boars (Lanada et al., 2005). In Western Kenya, the standard payment for boar services is a weaned piglet, which is given after a successful mating results in weaned offspring. This observation is consistent with findings from the Philippines published by Lanada et al. (2005).

To curb these boar-related problems, farmers started becoming more innovative.

Commercial pig farmers have over the years resorted to training some of their team members specifically to be able to handle these boars especially during heat checking, feeding, vaccination, etc. Also, taming the boars was a common practice. Using walk boards while walking the boars was also resorted to as a way of preventing or minimizing sustained injuries. Sadly, these methods did not give the best results as some team members still sustained varied levels of injuries while working with boars. These led to a decrease in the productivity of the farms. To ensure sustainable productivity, John McGlone in 2016 developed a novel patented boar pheromone, BOARBETTER® to replace the need for a live boar in a swine farm. BOARBETTER® pheromone is a natural pheromone that contains Boar Saliva Analog (BSA). It is the patented triple boar saliva pheromone that is a combination of three molecules (Androstenone, Androstenol and Quinoline). It is a water-based product containing analogs of the natural pheromone molecules. The active ingredients in BB are swine pheromones: 0.006% and inert ingredients: 99.994% (McGlone, 2016). McGlone et al. (2019) found that not one, but three boar-specific volatile molecules were unique in boar saliva (androstenol, androstenone and quinoline) compared to the saliva of sows. The combination of these three boar-specific molecules induced more complete sow sexual behavioral expressions, than the mixture of only androstenol and androstenone (McGlone, 2016). The pheromone is sprayed (4 ml) at the snout of the sow and the back pressure test is done almost immediately by a trained heat checker. This has shown to be more beneficial than using the teaser boars.

Internalizing foreign technology and farmers enlightenment will in no small way revamp the livestock sector of the country which is in the doldrums and reposition it for greater productivity and profitability. Most farmers are unaware of advances made in animal production, thereby sticking to old production practices which raises their cost of production and diminish their profit margin.

Sow performance is usually a measure of the number of piglets farrowed and weaned per sow per reproductive cycle. The number of litters a pig is able to farrow is very important in any pig enterprise. To have higher litter size, a good knowledge of heat/estrus detection is critical. The objective of this study was to do a comparative analysis of the performance of sows under BB and teaser boars in commercial pig farms and draw cost analysis of the duo. We hypothesized that use of BB would improve performance and reduce costs incurred in pig production.

Materials and methods

3.1 Location and duration of the study

This study was carried out at Country View, Livingston's Enterprises Inc., Fairbury, Nebraska, USA. Country View is located at 56308 720th Rd, Fairbury, NE 68352, USA with a latitude of 40.26354 (N $40^0$15'48.73212") and a longitude of -97.27277 (W $97^0$16'21.9612"). Fairbury records 32 inches of rain and 25 inches of snow, on average, per year. The US average is 38 inches of rain and 28 inches of snow per year. In Fairbury, temperatures typically vary from -7^0C to 32^0C and is rarely below -17^0C or above 37^0C; the summers are hot, humid, wet and mostly clear and the winters are freezing, dry, windy and partly cloudy. The study lasted for 32 weeks.

3.2 Experimental animals

A total of 4,810 F_1 (American yorkshire [sow] X landrace [boar]) weaned sows were used for this field trial. These sows were selected based on the following 5 criteria: (1) Lactation length (milked for 21 days, with good reserves of milk); (2) Good body condition (the sows had no shoulder sores, prolapse, showing no sign of disease, etc.); (3) Litter size (the sow farrowed at least 14 piglets from her previous litter); (4) Weaning size (the sow weaned at least 10 piglets from her previous litter); and (5) Parity (the sows that were used were selected from parities 1-6). After weaning, weaned sows were taken to the breeding section of the farm.

3.3 Experimental diets

Tables 1, 2 and 3 represent the different experimental diets used during this field trial. Feed was rationed based on the stage of gestation of the animal as follows:

- Gestation diet (Table 1): Gestation diet served triple purpose: it was given to sows on BB, sows on teaser boars and teaser boars. They received 3.63 kg of feed daily from weaning until five weeks after service. Their feed was reduced to 2.27 kg of feed daily from week 6 after service until 14 weeks after service (two weeks prior to farrowing).
- Lactation diet (Table 2): At 15 weeks, their feed was adjusted back to 3.63 kg daily for both sow groups, this time, on a lactation diet. This was to prepare the dam for the lactation coming her way. They were maintained on this ration until their farrowing day when their ration was increased to 14.52 kg (fed four times daily). They remained on 14.52 kg of feed until weaning.
- Creep feed (Table 3): This is the solid feed that's gradually introduced to the piglets one week after farrowing. They were maintained on this feed, from day 8 of age, until weaning. They were fed ad libitum. Water was given ad libitum to the animals throughout the experimental period.

Table 1. Feed composition of the gestation diet used in this field experiment

Feed ingredients	Quantity (kg)
Corn – Coarse Rolled	605.20
Corn – Micro Flush	2.80
DDGS	323.22
SBM – Hi Pro	30.00
Calcium Carbonate	15.00
Monocal	7.00
Biolys	4.65
Mixing Salt	5.00
Biotin 100	1.50
LEI Sow VTM 3#	1.50
Threonine	0.75
Choline Chloride	1.25
Optiphos 2000	0.50
Engage-M	0.50
Tryptophan	0.15

Pot Chloride 50	0.24
Vit E 20000	0.24
Visano-Sow	0.50
	1,000.00 kg

Table 2. Feed composition of the lactation diet used in this field experiment

Feed ingredients	Amount (kg)
Biolys	5.45
Biotin 100	1.50
Corn – Micro Flush	2.50
Calcium Carbonate	12.00
Choline Chloride	1.25
Corn – Course Rolled	628.54
Corn oil	30.00
DDGS	60.00
Engage-M	0.50
LEI Sow VTM 3#	1.50
Luvecell	0.50
Methionine	1.32
Mixing salt	5.00
Monocal	14.50
Optiphos 2000	0.50
SBM – Hi Pro	232.60
Threonine	1.60
Tryptophan	0.24
Visano	0.50
	1,000.00 kg

Table 3. Feed composition of the creep feed used in this field experiment

Feed ingredients	Amount (kg)
SBM – Hi Pro	62.50
Calcium Carbonate	10.50
Mixing salt	4.50
Biolys	6.17
Threonine	1.20
Methionine	0.24
Tryptophan	0.35
Optiphos 2000	0.15
Corn – Micro Flush	2.50
Skycis	0.15
Int. C	0.24
LEI GF VTM 3#	1.50
Corn – Coarse rolled	785.00
DDGS	125.00
	1,000.00 kg

3.4 Data collection

Data were collected on:

a. Sow performance
b. Piglet performance.
c. Cost effectiveness was calculated.

3.5 Sourcing of semen and post-cervical artificial insemination (PCAI) straws

Live Duroc semen (terminal line) was supplied to Country View, Livingston's Enterprises Inc., by DNA Swine Genetics throughout the study period. The DNA Genetics Line 600 Duroc is the most widely used terminal boar line in North America. The PCAI insemination straws were supplied by DNA Swine Genetics. DNA Swine Genetics is located at 4438 Old Mill Court, Columbus, Nebraska 68601.

3.6 Breeding method used

The PCAI (post-cervical artificial insemination) method of breeding was used during this field study and not the conventional artificial insemination method. 12 ml of semen was used per breeding. 1 ml of semen contained 4.125 million spermatozoa (minimum).

3.7 Sourcing of BOARBETTER®

BOARBETTER was supplied to Country View, Livingston's Enterprises Inc., by Animal Biotech (a laboratory specialized in animal welfare) throughout the study period. BOARBETTER is a patented product of Animal Biotech in partnership with Texas Tech University.

3.8 Sourcing of other materials used

Other materials used during this study include:

- Livestock spray paint: this is an aerosol spray paint which was used in marking or identifying sows on estrus. It was made in Denmark and supplied by DNA Swine Genetics.
- Insemination bags and gun: these were very useful in making insemination efficient. The semen delivered comes in 500 ml bag. This insemination bag houses the semen bag and helps to maintain the temperature of the semen to remain viable. The insemination gun was used to send the semen into the semen straws.
- Ultrasound scanner: a portable ultrasound scanner (Esaote Mylab Delta, Chagrin Falls, OH, USA) was used for pregnancy checking/detection. This was done five weeks after first service to confirm pregnancy. Ultrasound images were collected at a frequency of 8.6 MHz This ultrasound scanner was supplied by Valley Vet, Kansas State.
- Ultrasound gel: this is a gel used when carrying out heat checking. It is normally applied on the surface of the nub which is used in rubbing the abdomen of the sow while focusing on the screen of the ultrasound scanner to see the implanted embryos on the uterine walls. This gel was supplied by Valley Vet, Kansas State.
- Walking boards: these boards were used to protect the personnel handling the movement of the boars. They were purchased from Valley Vet, Kansas State.
- Safety boots, nose masks and ear plugs: these were vital materials for safety in hog facilities. They were purchased from Walmart, Fairbury, Nebraska.

3.9 Vaccination of the experimental animals

The experimental animals were vaccinated strictly following the farm's vaccination schedule under the supervision of a licensed Swine Veterinarian. This schedule was made available by a licensed Swine Veterinarian belonging to the American Association of Swine Veterinarians.

3.10 Pregnancy checking

Pregnancy checking is a method employed to ascertain the rate of conception after breeding. Oftentimes, sows that couldn't conceive after service/breeding recycle after 21 days. Pregnancy checking is done after 5 weeks following breeding/insemination with an ultrasound scanner for pigs.

3.11 Feed conversion ratio (FCR)

FCR was calculated using:

$$FCR = \frac{\text{feed intake}}{\text{body weight gain}}$$

Where:

FCR = feed conversion ratio;

Feed intake (measured in kg) and

Body weight gain (measured in kg).

3.12 Experimental design

Weaned sows were heat checked on an alternative basis. On two weeks interval, some sows were heat checked using teaser boar exposure while others were heat checked using pheromone. Therefore, the experimental animals were grouped into two groups:

- groups that were heat checked using teaser boar exposure (serving as the control group – 2280 sows), and
- groups that were heat checked using pheromone (serving as the treatment group – 2530 sows).

The experiment was executed in t-test.

$$t = \frac{Mx - My}{\sqrt{\left[\frac{\left(\sum x^2 - \frac{(\sum x)^2}{Nx}\right) + \left(\sum Y^2 - \frac{(\sum Y)^2}{Ny}\right)}{Nx + Ny - 2}\right] - \left[\frac{1}{Nx} + \frac{1}{Ny}\right]}}$$

Where:

t = student's t-test

Σ = sum of the following scores

M_X = mean teaser boars

M_Y = mean for pheromone

X = score in teaser boars

Y = score in pheromone

N_X = number of scores in teaser boars

N_Y = number of scores in pheromone

3.13 Statistical analysis

The data from the field trial was analyzed using Statistical Package for Social Sciences (SPSS) version 20.

Results

The results on the performance of piglets arising from sows treated with pheromone**,** cost effectiveness of pheromone versus teaser boars and cost of feeding 40 teaser boars are shown in Tables 4, 5 and 6, while the cost effectiveness of using teaser boars and pheromone are summarized in Figure 1.

Table 4. Performance of piglets arising from sows treated with pheromone

Treatments/Parameters	Control	Pheromone	*P*
Weight at birth (kg)	0.68±0.13[a]	0.19±0.13[b]	0.02
Weight at weaning (kg)	3.18±0.21[b]	4.54±0.26[a]	0.00
Piglets weaned per sow	10.50±0.02[b]	15.58±0.05[a]	0.00
Weight gain (kg)	2.50±0.13[b]	3.63±0.21[a]	0.00
Total creep feed intake (kg)	12.70±0.00	12.70±0.00	NS
FCR (based on creep feed intake)	5.08±0.02[a]	3.50±0.12[b]	0.00

[ab]means on the same row with different superscripts are significantly ($p<0.05$) different. SEM=standard error of mean. NS=Not Significant. FCR=feed conversion ratio.

There were significant ($p<0.05$) differences in weight at birth, weight at weaning, weight gained and feed conversion ratio (FCR). There was no significant ($p>0.05$) difference in total creep feed intake.

Table 5: Cost effectiveness of pheromone versus teaser boars

Treatments/Parameters	Control	Pheromone
Total sows served	2,280.00	2,530.00
Volume of pheromone sprayed per sow daily (ml)	0.00	4.00
Total number of times pheromone was sprayed	0.00	6,325.00*
Total volume of pheromone used (ml)	0.00	25,300.00
Volume of pheromone in a bottle (ml)	0.00	500.00
Cost of a bottle of pheromone ($)	0.00	$500.00 (₦382,396.65)
Cost of 51 bottles of pheromone used ($)	0.00	$25,500.00 (₦19,502,229.20)

***Half of the sows used in the pheromone group were served three times. Others in same group were served twice.*
$1= ₦764.7933.00.

Table 6: The cost of feeding the 40 teaser boars

Treatments/Parameters	Control	Pheromone
Amount of feed (kg) consumed daily by a teaser boar	3.63	0.00
Amount of feed (kg) consumed daily by 40 teaser boars	145.15	0.00
Amount of feed (kg) consumed weekly by 40 teaser boars	1,016.05	0.00
Amount of feed (kg) consumed by40 teaser boars for 8 weeks	8,128.38	0.00
Cost of a kilogram of feed ($)	11.10 (₦5,128.20)	0.00
Cost of 8,128.38 kilograms of feed ($)	90,225.00 (₦41,683,950.00)	0.00

$1 = ₦764.7933.00.

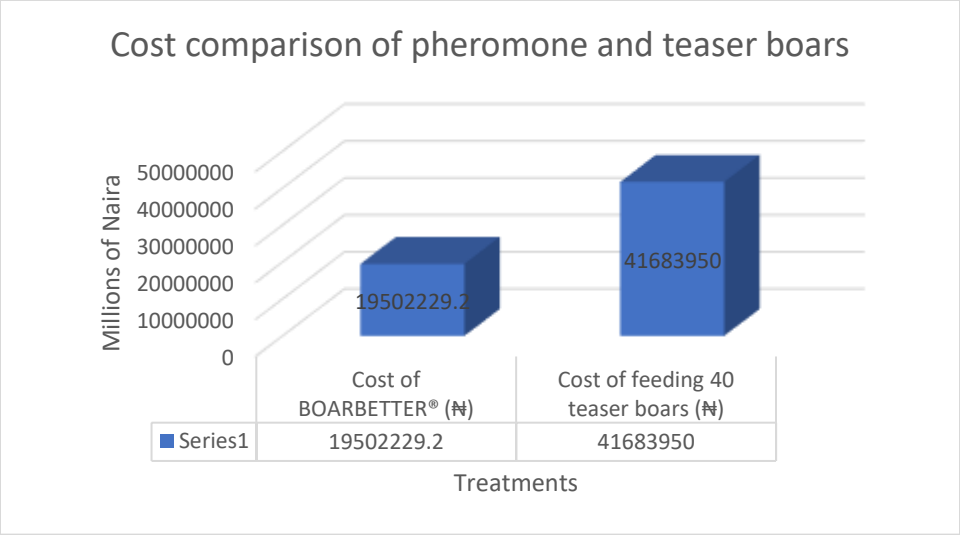

Figure 1: Cost effectiveness of using teaser boars and pheromone

Discussion

Performance of piglets arising from sows treated with pheromone

Weight at birth was higher in piglets in control (0.68 kg) compared to piglets in pheromone (0.19 kg). Weight at weaning was higher in piglets in pheromone (4.54 kg) compared to piglets in control (3.18 kg). Piglets weaned per sow was higher in piglets in pheromone (15.58) compared to piglets in control (10.50).

Weight gain was higher in piglets in pheromone (3.63 kg) compared to piglets in control (2.50 kg). FCR based on feed intake was higher in piglets in control (5.08) compared to piglets in pheromone (3.50).

The variations in weight at birth of piglets from sows under control and pheromone conditions may suggest that this trait had been affected by the breeding methods under consideration. Even though higher weight at birth is desired by farmers, it may lead incidences of overfat fetuses and possible dystocia which if not well managed can culminate into serious complications and possible loss of the dam. This may imply that pheromone has impressive effects on pregnancy in swine. This will have untold negative consequences on the farmer.

Piglets born to sows under pheromone reached better weaning weight than their counterparts in control. It may go a long way to explain that pheromone has longer and more positive effects, not only on the sows, but even on the offspring. Notwithstanding the average weights at birth, pheromone piglets outgrew the control piglets.

The long-lasting effect of pheromone on sows is also evident in the number of piglets weaned per sow per litter. Sows that received the treatment weaned more piglets compared to sows on the control group, evidence of superiority of pheromone to teaser boars in this regard. This is also in line with the findings of McGlone (2016) who recorded more piglets in sows that received pheromone than in sows in control.

The average number of piglets weaned per sow in this study was higher than that reported by Sinha et al. (2015). This high number may be directly attributed to low pre-weaning piglet mortality in both groups in this study. Secondly, the kind of breeding technique used can have an impact on pig farming productivity and reproducibility.

In pig production, artificial insemination is the preferred breeding method because it reduces the likelihood of breeding estrous sows or gilts with infertile boars (Njoga et al., 2021); this, in turn, reduces the likelihood of post-breeding conception failure or repeat breeder syndrome (Ate and Oyedipe, 2011). Interestingly, this study was carried out using artificial insemination because of the availability of facilities and technical know-how needed for the technique. Conversely, the open mating and hand breeding practices that are frequently used in farms (Njoga et al., 2021) may actually make conception more difficult because they don't stop infertile boars from reproducing (Nauta et al., 2001). When the dominant boar is infertile, it could make things worse by preventing subordinate males who are fertile from reproducing (Nauta et al., 2001; Horback and Parsons, 2018). As a result, there might be fewer farrowing and weaned piglets per sow per litter and a lower conception rate in the piggery.

The weight gain and feed conversion ratio of the piglets arising from sows treated with pheromone were better compared to the control group. Probably, the pheromone received by the sows altered the level and the composition of milks after farrowing. Since the two groups of piglets received same amount of creep feed and all things being equal, it can be suggested that the higher weight gain as seen on the treated group was as a result of improved milk utilization on the piglets. It’s possible that the pheromone received could have altered the micro-organisms inhabiting the gut of the piglets thereby leaving more beneficial microbes on the gut. With this, the piglets from the treatment group made more efficient utilization of the creep feed resulting to lower FCR.

Economics of using pheromone

A total sum of ₦19,502,229.20 was used to procure pheromone for heat detection of 2,530 sows to produce 15.58 piglets per sow at a weaning weight of 4.54 kg, while a whooping sum of ₦41,683,950 was used to feed 40 teaser boars for heat detection of 2,280 sows to produce 10.50 piglets per sow at a weaning weight of 3.18 kg. This implies that pheromone is almost four times more efficient than teaser boars in swine reproduction.

While labour costs were not measured, there appears to be some labour savings when using pheromone as against when using an adult live boar. This saved the time that was required to get the boars out of the pen and move them throughout the barn. It also contributed to reducing the risk of human injury by boars. Boar handling requires trained personnel and hiring these persons increases the cost of running a commercial swine farm. The use of pheromone may eventually eliminate the use of live boars in the sow barns, which would save costs and reduce the risk of injury to people.

Conclusion

In conclusion, we can deduce the following:

- Pheromone was twice more efficient than teaser boars in heat detection of sows.
- Piglets under pheromone had better growth performance.
- More piglets were weaned under pheromone.
- Pheromone reduces accidents in pig breeding to the barest minimum.
- Pheromone removes cost of personnel training required in the use of teaser boars.
- Pheromone reduces time required for breeding/mating of sows.

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