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Preface

We would like to present, with great pleasure, the inaugural volume-7, Issue-3, March 2021, of a scholarly journal, *International Journal of Environmental & Agriculture Research*. This journal is part of the AD Publications series *in the field of Environmental & Agriculture Research Development*, and is devoted to the gamut of Environmental & Agriculture issues, from theoretical aspects to application-dependent studies and the validation of emerging technologies.

This journal was envisioned and founded to represent the growing needs of Environmental & Agriculture as an emerging and increasingly vital field, now widely recognized as an integral part of scientific and technical investigations. Its mission is to become a voice of the Environmental & Agriculture community, addressing researchers and practitioners in below areas.

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Environmental science and regulation, Ecotoxicology, Environmental health issues, Atmosphere and climate, Terrestrial ecosystems, Aquatic ecosystems, Energy and environment, Marine research, Biodiversity, Pharmaceuticals in the environment, Genetically modified organisms, Biotechnology, Risk assessment, Environment society, Agricultural engineering, Animal science, Agronomy, including plant science, theoretical production ecology, horticulture, plant, breeding, plant fertilization, soil science and all field related to Environmental Research.

Agriculture Research:

Agriculture, Biological engineering, including genetic engineering, microbiology, Environmental impacts of agriculture, forestry, Food science, Husbandry, Irrigation and water management, Land use, Waste management and all fields related to Agriculture.

Each article in this issue provides an example of a concrete industrial application or a case study of the presented methodology to amplify the impact of the contribution. We are very thankful to everybody within that community who supported the idea of creating a new Research with *IJOEAR*. We are certain that this issue will be followed by many others, reporting new developments in the Environment and Agriculture Research Science field. This issue would not have been possible without the great support of the Reviewer, Editorial Board members and also with our Advisory Board Members, and we would like to express our sincere thanks to all of them. We would also like to express our gratitude to the editorial staff of AD Publications, who supported us at every stage of the project. It is our hope that this fine collection of articles will be a valuable resource for *IJOEAR* readers and will stimulate further research into the vibrant area of Environmental & Agriculture Research.



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Determinants of Farmer's Participation in Soil and Water Conservation Practices in North- Central Highlands of Ethiopia

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Abstract— Soil erosion is the major threat in the highlands of Ethiopia. Even if large number of conservation campaigns have been undertaken, the efforts are less effective, because of low intention to farmer's participation in soil and water conservation (SWC) works. The objective of this study was to identify determinants of farmer's participation in soil and water conservation (SWC) in Borena woreda, north-central highlands of Ethiopia. Samples of 148 households were selected following Stratified, proportional sampling technique. Household survey, focus group discussion, and key informant interview methods were used to collect primary data. Secondary data were also collected from Woreda agriculture office, published, and unpublished documents. Both descriptive statistics and Binary logistic regression model using Statistical Package for Social Science (SPSS) version 20 were used to analyze the data. About 76.35% of the respondents were participants. However, 57.52% of participants were without their interest. The binary logistic regression results showed that perception, extension service, training, and slope of their land found to have a positive and significant influence on farmer's participation. Whereas, age of household head and off-farm income found to have negative and significant influence on farmer's participation in SWC practices. When farmers get extension and training service, they become willing to participate in SWC practice because they acquire necessary information and skill. Therefore, the Woreda Agricultural office should provide extension service and encourage farmers training for the sustainable management of the land and its productivity.

Keywords— Binary logit model, Borena Woreda, Determinants, Extent, Farmer's participation, SWC.

I. INTRODUCTION

Ethiopia depends on an agricultural-based economy with a rapidly increasing population, soil erosion, and sedimentation problems caused by cropland expansion, overgrazing, and over-exploitation of forests (Biratu and Asmamaw, 2016). Soil erosion is a severe phenomenon in Ethiopian highlands due to inappropriate cultivation on steep farmland (Abate, 2011). Studies showed that 50% of the highlands have significant soil erosion, 25% of it is highly eroded and 4% of the eroded land is beyond reclamation (Akalu et al., 2016). After the emergence of famine and drought in the 1970s, the government of Ethiopia launched various soil and water conservation measures in various parts of the country (Teshahunegn et al., 2012). A large number of structural, biological, and agronomic soil and water conservation technologies which include, soil and stone bunds, grass strip, agroforestry techniques, and water harvesting options such as tied ridges and pond construction have been undertaken throughout the country (Shiferaw et al., 2007). Even if a large number of conservation campaigns were undertaken through Food-For-Work (FFW) programs, the efforts didn't bring significant changes as expected (Kidane et al., 2014; Teshome et al., 2016). Traditional agricultural practices and improper design of soil and water conservation (SWC) practices increase the severity of soil erosion (Abate, 2011). Moreover, the farmers have given little attention to decisions making and participation in most processes of soil and water conservation activities (Amsalu and Graaff, 2007). Having these problems at hand, there is no research conducted in this area related to farmer's participation in soil and water conservation practices. Hence, this study aimed to identify determinants of farmer's participation in soil and water conservation practices in the north-central highlands of Ethiopia.

II. RESEARCH METHODS

2.1 Description of the Study Area

The study area is located in South Wollo zone, Amhara region, north-central Ethiopia (figure1). It is geographically located between 10° 32' N to 10° 56' N latitude and 38° 32' E to 38° 56' E longitudes. Its altitude ranges from 500 meters above sea level to 3600 meters above sea level. The dominant soil types in the study area are leptosols, cambisols, andosol, and vertisols. The distribution of rainfall is bimodal, which occurs from June to September, locally known as "Kiremt", and March to May, which is locally known as "Belg". The mean annual rainfall depth varies from 900 to 1200 mm per year. The mean annual temperature of the area varies from 16.25°C to 25°C. The farming system is mixed farming; cereal crop cultivation and livestock rearing. The major crops grown in the area include teff (*Eragrostis*), wheat (*Triticum Vulgare*), barley (*Hordeum vulgare*), maize (*Zea mays*), field pea (*Pisum sativa*), and linseed (*Linum usitatissimum*).

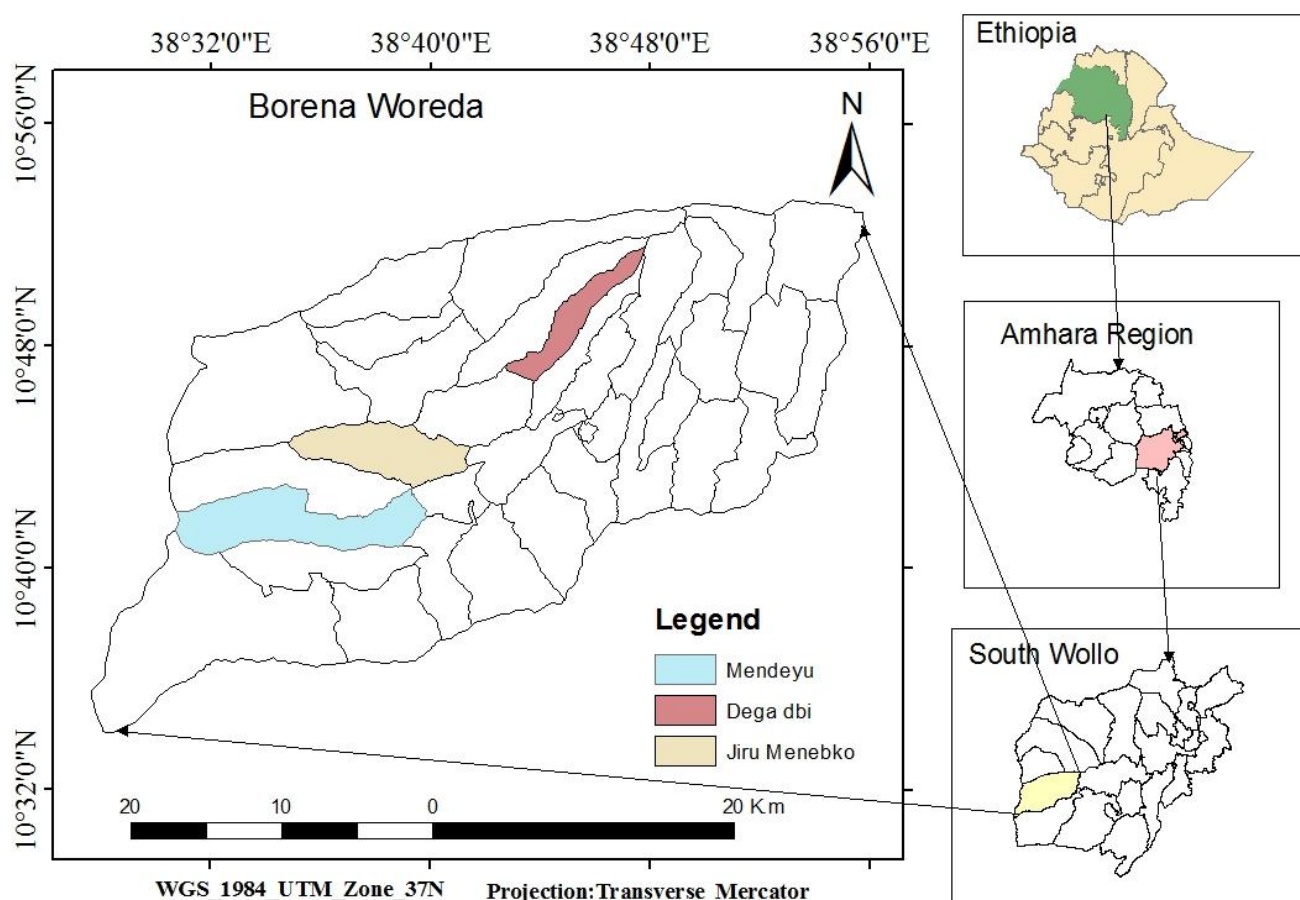


FIGURE 1: Map of the study area

2.2 Sampling techniques

Stratified sampling techniques were used to select sample kebeles and respondent farmers. Sample kebeles from Borena woreda kebeles were selected purposively based on their agro-ecological location and abundance of SWC practices. So three representative kebeles (Mendeyu, Menebuko, and Dega Dbi) were selected from Kolla, Woina Dega and Dega agro-ecological zones respectively. To identify determinants of farmer's participation in SWC, the sample households were selected from each selected kebele proportionally. The sample size was determined using a formula adapted in Israel (1992).

$$n = N / (1 + N(e^2))$$

Where: n is the sample size, N is the population size, and e is the level of precision (8%). Accordingly, from 2860 household heads 148 sample households were selected from the three kebeles.

2.3 Data collection and Analysis

For this study, both primary and secondary data sources were used. To get detailed information concerning the farmer's participation in SWC, the primary data were collected through a household survey, focus group discussions, and key informant interviews. The secondary data were also collected from different written materials at Woreda and kebeles about population, and soil and water conservation. Simple descriptive statistics was used to analyze the extent of farmer's participation. Binary Logistic regression analysis was employed to determine which hypothesized independent variables significantly affect farmer's participation in soil and water conservation practices in the study area because the dependent variable of this study is dichotomous.

2.4 Model specification

The use of binary logistic regression model gives maximum likelihood estimates to overcome most of the problems associated with linear probability models and provide parameter estimates that are asymptotically consistent and efficient. The dependent variable is dichotomous, where the response y_i is binary assuming only two values that can be coded as one or zero for convenience. This could be defining as:

$$y_i = \begin{cases} 1, & \text{if the } i^{\text{th}} \text{ household is participant in SWC activities} \\ 0, & \text{if not participant.} \end{cases}$$

Here, y_i take value one and zero with probability of p_i and $1-p_i$ respectively.

The probability that the i^{th} household to participate in SWC activity is;

$$p_i = \frac{1}{1+e^{-Z_i}} \quad (1)$$

Therefore, the odds ratio of the probability of households participating in SWC activities to probability of households not participating in SWC activities can be written as:

$$\text{Odds Ratio} = \frac{p_i}{1-p_i} = e^{-Z_i} \quad (2)$$

Taking its natural logarithm, we got the log of the odds ratio, which is known as logit model.

$$\ln\left(\frac{p_i}{1-p_i}\right) = Z_i \quad (3)$$

If the error term (ε) is taken in to account, the logit model becomes:

$$Z_i = \beta_0 + \beta_1 X_{1i} + \beta_2 X_{2i} + \dots + \beta_n X_{ni} + \varepsilon_0 \quad (4)$$

Where, β_0 is the intercept, which tells us the log odds in favor of participating in SWC practices when the coefficient of all included explanatory variables are assumed zero. $\beta_1, \beta_2, \dots, \beta_n$ are slope parameters to be estimated in the model. The slope tells how the log odds in favor of participating in SWC practices change as each independent variable change. Z_i tells as the logs of the odds ratio in favor of participating in SWC practices. In this study, the above econometric model (equation 5) is to identify factors affecting the participation of farmers in SWC practices. Z_i is a linear function of n explanatory variable (X_i) and expressed as:

$$Z_i = \beta_0 + \beta_1 EXTE_i + \beta_2 DIST_i + \beta_3 AGE_i + \beta_4 PERCEP_i + \beta_5 AREA_i + \beta_6 EDU_i + \beta_7 SLOPE_i + \beta_8 SEX_i + \beta_9 FAMSIZE_i + \beta_{10} SOILFERT_i + \beta_{11} TLU + \beta_{12} TRAIN_i + \beta_{13} OFF FARM + \varepsilon_i$$

Variance Inflation Factor (VIF) and Contingency Coefficient (CC) checked the occurrence multicollinearity problem with continuous and dummy explanatory variables respectively. So the value of VIF was less than 10 for continuous. For dummy variables, the value of contingency coefficient was less than 0.75. Therefore, there is no multicollinearity problem.

TABLE 1
DEFINITION AND UNITS OF MEASUREMENT OF THE EXPLANATORY VARIABLES.

Variables	Definition and units of measurements
EXT	Extension service (1= obtained and 0=not obtained)
DIST	Distance from residence to the farm land in minute or hour
AGE	Age of household head in years
PERCEP	Household head's perception on erosion problem (1=know 0=don't know)
AREA	Area of land (in hectare)
EDU	Educational status of household head (1=literate, 0=illiterate)
SLOPE	Slop of the plot (1= flat and 0 =otherwise)
SEX	Sex of household head (1=Female, 2=Male)
FAMSIZE	Number of family member
SOILFERT	Fertility status of the soil (1=fertile, 0= infertile)
TLU	Respondent's owned livestock (in tropical livestock unit)
TRAIN	Household head's taken training (1= yes, 0 =no)
OFF_FARM	Respondent's off farm income (1=have, 0= not have)

III. RESULTS AND DISCUSSIONS

3.1 The extent of farmer's participation in SWC practices

About 76.35% of the respondents were participants. However, 57.52% of participants were without their interest because they perceived that SWC measures implemented by the public campaign are less effective to combat soil erosion. From the focus group discussion, farmers also participated to be free from any penalty of being absent. Similarly, Wagayehu, and Sterk (2002) reported that the majority of the households participated simply because they were forced to do by the village administration and the experts. Because farmers assumed that conservation is not for the sake of protecting the soil and water from erosion rather to meet the demands of the government's five-year development program. Abrham et al (2016) also reported that farmers were reluctant to participate because of poor effectiveness of conservation measures and unfair selection of farmers for layout and design activities. The majority (68.14%) of respondents participated at the implementation stage (Table 2). This is in line with Abrham et al (2016) that almost three fourth of farmer's participated in the implementation phase in both watersheds.

TABLE 2
THE EXTENT OF FARMER'S PARTICIPATION IN SWC ACTIVITIES

Variables related to SWC	Category	Frequency	(%)
Participation	Participant	113	76.35
	Non Participant	35	23.65
Way of participation	Voluntarily	48	42.48
	Forced	65	57.52
Stages of participation	Planning	13	11.51
	Implementation	77	68.14
	After implementation	23	20.35

Note: Way of participation and stages of participation analyzed from participants

3.2 Determinants of farmer's participation in soil and water conservation practices

3.2.1 Age of household head

It was negatively related to farmer's participation in SWC practices and statistically significant at 1% significant level ($\beta = -0.793$; p -value = 0.000). The negative sign indicates that as the age of farmer increases, the probability of participating in SWC practices decreases. The odds ratio also showed that a one-year increase in the age of household head decreases their participation in SWC practices by a factor of 1.104. As SWC practices require more energy, older farmers have no enough energy to do. Due to this, they give their farmland to younger farmers as rent. This result was in line with (Belachew et al.,

2020; Daniel and Mulugeta, 2017; and Mohammed et al., 2018) who reported a negative correlation between age and adoption of introduced SWC measures. This is because old farmers become exhausted and unable to give care for their farmlands. In contrary to this, Amsalu and Graaff (2007) indicated a positive correlation between age and willingness to participate in SWC programs due to the longer year farming experience of older farmers. Fikru (2009) also reported that older farmers were more aware of the problems of erosion and the importance of soil and water conservation practices.

3.2.2 The slope of farmland

The slope of the land was positively correlated with farmer's participation in SWC practices and significant at a 1% significant level ($\beta = 1.480$; p -value = 0.004). The result showed that as the land steepness increase by 1%, their participation in SWC practices also increases by a factor of 2.646. Farmers having steep slope fields perceive that the presence of erosion problem. Therefore, farmers having a steep slope of land actively participate in SWC practices because of its difficulty in the absence of public campaign work. Similarly, Belete (2018) reported that the slope of a farm plot found to be statistically significant and positively correlated with continued use of structures at less than 5% probability level. A household inclines implementation of soil and water conservation structures as he or she owns very steep land that could be exposed to soil erosion. This implies that households farming steep land are more likely to adopt conservation structures than less steep lands. Likewise, Desalew and Aklilu (2017) reported that the slope of a plot was an important variable that positively affecting the probability of using SWC technologies at a 5% significance level. Farmers were tending to invest in conservation practices where their farm plots are located on higher slopes. Due to more visibility of soil erosion problems in steeper slopes, farmers could use conservation measures. On the contrary, Kessler (2006) states that more sloping fields do not influence farmer's decision to the adoption of SWC.

3.2.3 Extension contact of the household head

It was positively correlated (consistent with the hypothesis) with farmers' participation in SWC practices; and significant at 1% significant level ($\beta = 0.774$; p -value = 0.003). The odds ratio indicated that in each contact of farmers with extension workers, farmers' participation in SWC practices increased by a factor of 1.811. Because when farmers frequently communicate with extension workers, they can get the necessary information, acquire new skills and knowledge, and became more aware to use conservation practices. This result is in line with Derejaw et al (2013) who reported that frequent contact with development agents found to affect the use of improved soil conservation technologies positively and significantly at a 1% significant level. Every contact of the farmer with the development agents increased the probability of use of improved soil conservation technologies by 9.03%. In the same way, Desalew and Aklilu (2017) indicated that the effect of extension contact on farmer's perception was positive and significant at a 5% significant level. Farmers who have more contact with extension workers would acquire more information related to the benefit of SWC, the techniques of implementation, and maintenance. In contrary to this result, Amsalu and Graaff (2007) reported a negative correlation between extension contact and adoption of soil and water conservation. Farmers having contacts with extension agents tend to reduce their investments in SWC practices because the agricultural extension mainly focused on crops and livestock production.

3.2.4 Perception of farmers on the problem of soil erosion

It was positively correlated with farmer's participation in SWC practices; and significant at 1% significant level ($\beta = 2.253$; p -value = 0.000). The binary logistic regression result showed that the odds of farmers who perceive the problem of soil erosion was 1.847 times the odds of farmers who do not perceive. Because farmers who perceived about soil erosion problem on their farmland understand the severity of soil erosion and ultimate decline of productivity. Similarly, Adugnaw and Birhanu (2013) reported that perception on the problem of soil erosion had a positive and significant influence on the use of structural SWC measures. Likewise, Belete (2018) also indicated that farmers who feel that their farmlands are prone to soil erosion are more likely to adopt physical soil conservation measures than those who do not perceive the problem of soil erosion. From his result, the odds of a farmer who perceived soil erosion better to adopt conservation structure was 3.363 times the odds of farmers that did not perceive it. This implies farmers who feel that his/her farmland was prone to soil erosion were more likely to use SWC measures.

3.2.5 Training

Households obtained training was positively correlated with farmer's participation in SWC practices and significant at 1% significant level ($\beta = 1.916$; p -value = 0.001). The model result showed that the odds of a farmer who take training was 3.534 times the odds of farmers who do not take training. Farmers, who take training, acquire adequate and recent information and

technical support about the implementation of SWC mechanisms as well as their relevance. Therefore, they improve their awareness in the participation of SWC practices and facilitate the use of conservation measures. However, according to Belachew et al. (2020), training influenced the adoption of check dam negatively at a 5% significant level. Because when farmers have not accessed all the relevant information or information they obtained was incomplete, they cannot adopt SWC technologies.

3.2.6 Off-farm income of the household head

This variable related to farmers' participation in SWC practices negatively and significant at 1% significant level ($\beta = -2.597$; p -value = 0.000). The odds of farmers who engaged in off-farm income was 1.014 times the odds of farmers who did not engage in off-farm income activities. Farmers do not want to spend their time in farmland when they get off-farm income source because most of the off-farm activities such as masonry, carpentry, and trading are carried out far from their farm area. In line with Belete (2018) that farmers engaging in off-farm activities discourage a household not to participate in soil and water conservation activities. Thus, off-farm activities influence farmer's continuous use of soil and water conservation technologies negatively and significantly at less than 5% probability. It decreases the probability of using SWC technologies by a factor of 0.376. Daniel and Mulugeta (2017) also reported that household heads who are not engaged in off-farm activity adopt introduced SWC practices .084 times greater than those who are engaged in the off-farm activity. Because there is labor competition between off-farm activity and SWC practices which restrain farmers from involving in implementing and maintaining conservation practices on their farmlands. In the same way, Derejaw et al (2013) explained that participation in off-farm activities keeps the labor force needed for SWC away from the farm. Moreover, the short-term benefit obtained from off-farm works may obscure the benefits obtaining from investments in SWC. However, Kessler (2006) found that income from off-farm activities does not influence a household's decision to invest in SWC measures.

TABLE 3
BINARY LOGIT MODEL FOR DETERMINANTS OF FARMER'S PARTICIPATION IN SWC PRACTICES

Variables	Coefficient(β)	Std. err	Sig.	Odds ratio
AGE	-0.793	0.261	0.000***	1.104
SEX	-1.759	0.534	0.672	.007
EDU	-0.04	0.472	0.850	.893
FAMSIZE	0.515	0.061	0.747	4.358
DIST	0.219	1.053	0.771	1.869
AREA	0.59	0.951	0.453	5.390
SLOPE	1.48	0.746	.004***	2.646
SOIL FERT	-0.083	0.183	0.263	.789
PERCEP	2.253	0.621	0.000***	1.847
-TLU	-0.484	0.073	.655	.251
EXT	0.774	2.415	.003***	1.811
TRAIN	1.916	3.414	.001***	3.534
OFF_FARM	-2.597	3.472	.000***	1.014
Constant	-2.101	9.762	.000	.002

Nagelkerke (R^2) = 0.825; Log likelihood = -82.397;
Model chi-square = 87.775; Number of observation = 148;
*** means Significant at $p \leq 0.01$.

IV. CONCLUSIONS

The findings of this study revealed that even majority of respondents are participant, their participation was mainly at the implementation stage to be free from penalty of being absent. The participation of farmers in SWC practices in the study area is affected by their perception of erosion problem, extension services, training, and slope of their farmland positively. On the hand, age and off-farm income affected farmer's participation in SWC practices negatively. Therefore, strategies those focus on enhancing and strengthening of the interest and ability of farmers like training and extension on the sustainable management of land should be continuously implemented.

ABBREVIATIONS

GIS: Geographic Information System

SWC: Soil and Water Conservation

SPSS: Statistical Package for Social Science

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Study of the influence of a Bioabsorbent derived from Orange Peel on a filtering soil using seawater irrigation by capillarity

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Abstract— *The effect of rain on our planet has been the most important meteorological phenomena to be reproduced by humans. It has been vital for the hydration of the soil, making it possible for agriculture to prosper and progress. However, the great secret of irrigation is in the ground, in the water tables and aquifers that store and manage water, storing every drop of rain and distributing the water through the underground river basins, indirectly irrigating everything, from the mountain to the sea, making the cultivation of crops possible. This means that the type of soil is as important as the water supply.*

Irrigation for agriculture has always simulated rainfall; therefore, it has copied irrigation from above and has focused on the soil drainage capacity. From this point of view, saline water is not beneficial for this activity, but it may be the only source of irrigation water for arid regions, especially in developing countries, where there's a scarcity of water and the population is rapidly growing. Storing irrigation water for both agriculture and the increasing population is necessary for the developing country's prosperity.

The use of seawater applied to irrigation is not a new technique, there's evidence that proves that in 1719 the Sestao's Carmelite monks, located in Vizcaya, made use of this practice.

When considering the possibility of irrigation without desalination, always through capillarity systems, it is essential to consider some critical factors, such as the substrate of the ground, the distance of the water table, the salt composition of the seawater, chemical reactions of the ground with the salts or the drainage of the ground. Modifying any of these parameters can cause salinization effects, loss of humidity or desertification of the substrate, amongst others.

This study shows the influence of a bio absorbent obtained from the orange peel on the behaviour of a substrate based on silicon sand.

Keywords— *Desertification, Desalination, Reusable, Seawater Table, Bioabsorbent.*

I. INTRODUCTION

Desertification is the result of climatic changes and human activity, which reduces productivity and the value of natural resources in arid and semiarid conditions (Aubréville, A.; 1949). This is the international definition established by the convention of the United Nations against desertification, approved on 17th June of 1994, and from that day a date has been commemorated as the World Day to Combat Desertification and Drought.

The main causes for desertification are the weather, erosion, ecological factors such as the kind of soil and ecosystem, as well as human activity. Before desertification occurs, there is an erosion of the ground: when the wind removes the excess of soil and dust particles from the ground, the soil then loses the water table and the regeneration of the soil becomes very difficult (Stringer, L.C.; 2008). The low drainage of the soil, torrential rains or drought are other phenomena that cause desertification. Climatic change is a harmful phenomenon, but the most harmful is human activity: fires, logging, overexploitation of aquifers, intensive farming, with massive use of chemicals, and some forestry practices (mountain or forest crops), etc. (FAO 2000).

Worldwide it is considered that a volume above 1.000 m³ per inhabitant and year is normally more than necessary for domestic use, industrial and agricultural. In consequence, it is estimated that a watershed suffers a loss of hydric stress when the water availability per inhabitant is below 1.000 m³/year or when the quotient between water withdrawal and historical

annual average runoff is greater than 0.4. There are some basins of this type in North Africa, the Mediterranean region, the Middle and Near East, South Asia, North China, the United States of America, Mexico, Northeast Brazil, and the west coast of South America. The population of those who live in these basins is about 1.400 and 2.100 million people (Vörösmarty et al., 2000; Alcamo et al., 2003; Oki et al., 2003; Arnell, 2004).

Irrigation represents 70% of all the water extracted in the world and almost 40% is for agricultural production (Fischer et al., 2006). In fact, all the irrigated land represents 18% of all agricultural land in the world and produces 1.000 million tons of cereals each year, half of the total world supply. In fact, irrigated crops produce between two and three times more crops than those produced by rain alone. (Alexandratos, N., 2005).

In general, it seems that global warming will benefit the agriculture of developed countries in warm climates. On the contrary, the countries with a tropical or subtropical climate will suffer the effects of climatic change, and the dependence on imports from other countries will increase, as the difference between north and south, of alimentary security, will be accentuated (Canadell et al., 2007)..

Water management tends to improve the quality and quantity of available water regulating the use of water from the surface and underground, developing other sources of water, rationalizing water's consumption, or, controlling the contaminants and recovering them from wastewater through a depuration process. The objective of good water quality must be pursued in each river basin, so that measures relating to surface water and groundwater belonging to the same ecological, hydrological and hydrogeological system are coordinated (Directive 2000/60 / EC). From this point of view, the reuse of treated water is an essential element of the natural water cycle and, in fact, is seen as a measure to solve the problems of water scarcity.

Agriculture is the main cause of the lack of water around the world, 70% of all the water extracted and in some cases 95% in developing countries (FAO, 2017), is used for this activity, thereby reducing water availability for people.

It is expected that between now and year 2050, the demand for food will increase by 60% and the world's population will be around the 9.000 million (FAO, 2013). The actual and future situation will mean agriculture will need more and more water, and consequently lead to an increase in the global lack of water including those areas that currently have enough sources of water.

In order to reduce the agricultural pressure on the hydric sources, one of the possible solutions is the use of brackish or desalinated seawater. The advantage of the use of this type of water is that there are inexhaustible resources and they are not subject to climatic variations. So this type of water is strategically perfect to develop the availability of increased water resources for agricultural irrigation in water deficient areas. However, only the most advanced crops with higher economic margins can bear the costs of desalinated water (Martínez 2014). This desalinated or osmotized water is characterized by its low mineralization and significant imbalances in its composition. So it is not suitable for every type of supply (domestic, agricultural or industrial). In order to adapt the characteristics of the osmotized seawater to the requirements of the different uses, it must undergo post-remineralization treatments.

The depurated water reusability for crop irrigation is another option, and lately it has been used in many countries. Nevertheless, it is necessary to guarantee a minimum quality of water, as well as its functional uses depending on how it is treated.

On the other hand, another technique to address the lack of water and that hasn't been contemplated until now, is the use of seawater for the irrigation and development of crops.

The uses of capillary irrigation systems can offer a solution not only to the problem of water scarcity, but can also contribute substantially to the transformation of arid or desert areas into green areas and fields. It must be taken into account that the water rises in the soil due to evaporation and absorption by the roots of the plants. Water moves by capillarity (especially intense effect in arid climates) and by difference in humidity, (deeper ground levels remain wetter as they are protected, due to their distance from the soil surface, and from water losses due to evaporation and the absorption by plants). On the other hand, the water not only moves vertically, it also moves laterally. Therefore, it can be said that the water in the ground moves in any direction (Duchaufour 1978).

The use of seawater applied to irrigation is not a new technique, there's evidence that proves that in 1719 the Sestao's Carmelite monks, located in Vizcaya, made use of this practice. Later, on Oriñon beach (Cantabria) in 1959 a study was made of the use of seawater, without desalination, for irrigation, and achieving it without saturating the soil with salts (Esteban-Gómez, 1968).

Recently, there have been some successful investigations into the cultivation of chard using only seawater. One of the main characteristics is the low difference in height between the sea water and the crop (García et al., 2019), which is a good example of how to make a well-functioning irrigation system as well as reducing the costs. However, to successfully develop the cropping with higher yields, it is fundamental to optimize the substrate for the water and mineral supply of the crops. The tolerance conductivity of the soil will be a major parameter to control; otherwise, the salinity may affect the crops.

The objective of this study it to determine the influence of a bioabsorbent obtained from orange peel, on the behaviour of a substrate composed of silicon sand in order to reduce the salinity of seawater without a previous desalination, by applying this technique to capillarity irrigation.

II. MATERIAL AND METHOD

All the experiments for this project were carried out using capillary irrigation, which means that the seawater reached the substrates underground, and then rose through a tube.

Preliminary studies (García et al., 2019a) demonstrated that for irrigation it seawater can be successfully used, as long as it is supplied through the water table. However, this might not be enough, the soil is really important to optimizing irrigation with salty water. The kind of soil or substrate has to have particular characteristics (composition and granulometry) to reduce the salinity and store the humidity, in that way the crop can supply itself with water and nutrients as necessary, and at the same time, the salinity won't exceed the value limit to make crops grow. In all the previous experiments, the conductivity did not rise more than 2 mS/cm above 15cm ground level (García et al., 2019a). The fact that the soil can reduce the concentration of salts is possible because of the ion exchange between the water and the substrate. It is a dynamic process that develops on the surface of the particles, as a result of the electrical imbalances of the ground particles. In order to neutralize these charges, it adsorbs the ions from the solution of sand and sea water (Porta et al., 1994).

On this Project, silicon sand was used as a soil filter, as it is a material with great resistance to chemical compounds and water insolubility, and this is the reason why it is used as a swimming pool filter. The chosen granulometry was between 500-1000 μm , which has a very efficient drainage capacity and at the same time, enough capillary strength to absorb the seawater from the underground to the surface, because along the tube the silicon sand reduced the amount of conductivity.

A waterproofed channel with 80 cm long, 34.7 cm wide and 22 cm high was built with a drainage system to control the maximum height of the seawater. To facilitate water renewal a pump was used with a flow of 2L/day, working on an open water circuit. The aim was to create an artificial water table continually renewing the water and avoiding the formation of brines and an accumulation of salts from the salty water.

The technique used to check the ascension of the water was made using tubes of PVC, the diameter was about 57 mm, and so the section in contact with the seawater was of 25'51cm². The tubes were filled with the filtration samples and there were two types: the first one had silicon sand and the second type contained silicon sand with bioabsorbent at 5% w/w and a granulometry between 500-1000 μm . The samples with bioabsorbent could be divided in function by the physiochemical properties: there were samples with a bioabsorbent physically treated and another type that had a physical and chemical treatment. On Figure 1 it is shown a scheme of the samples and their contents. The total numbers of tubes used in the experiment were eighteen.

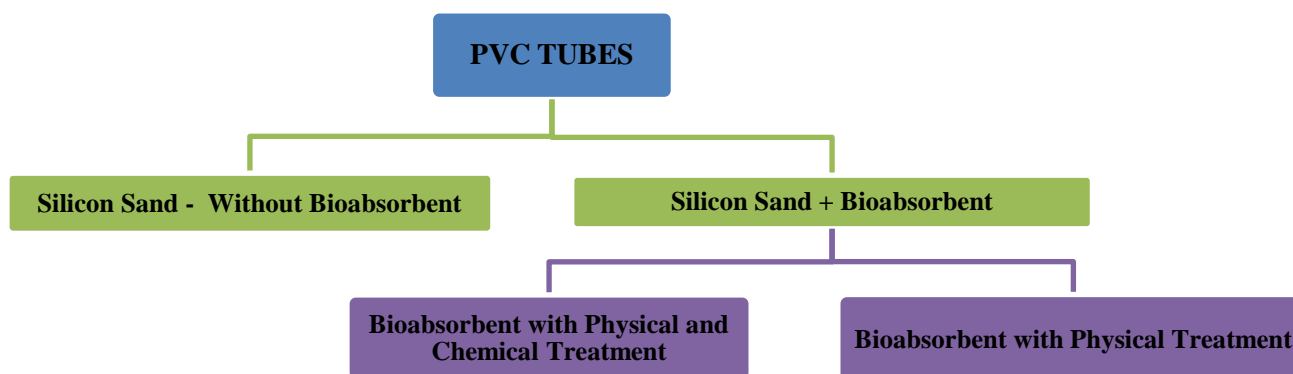


FIGURE 1: SCHEME OF THE TUBES AND TYPE OF SAMPLES

The first samples were extracted after sixteen weeks, and, at different heights, the values of pH, conductivity and humidity were measured using 9 tubes in total.

The remaining 9 tubes were subjected into a precipitation simulation. To do so, 500 mL of deionized water was added to the surface, and the samples were extracted at week eighteen, twenty and twenty-four, 2, 4 and 8 weeks after the water was added, respectively. The aim of this simulation was to check the behaviour of the ascension of the seawater through the variety of substrates with and without the bioabsorbent.

III. RESULTS

3.1 Results at week sixteen

Firstly, it can be appreciated that the seawater did not reach the surface of the samples that contained bioabsorbent, in addition, there appeared microorganisms (Figure 2). In contrast to the sample with silicon sand, in which the water reached the surface.

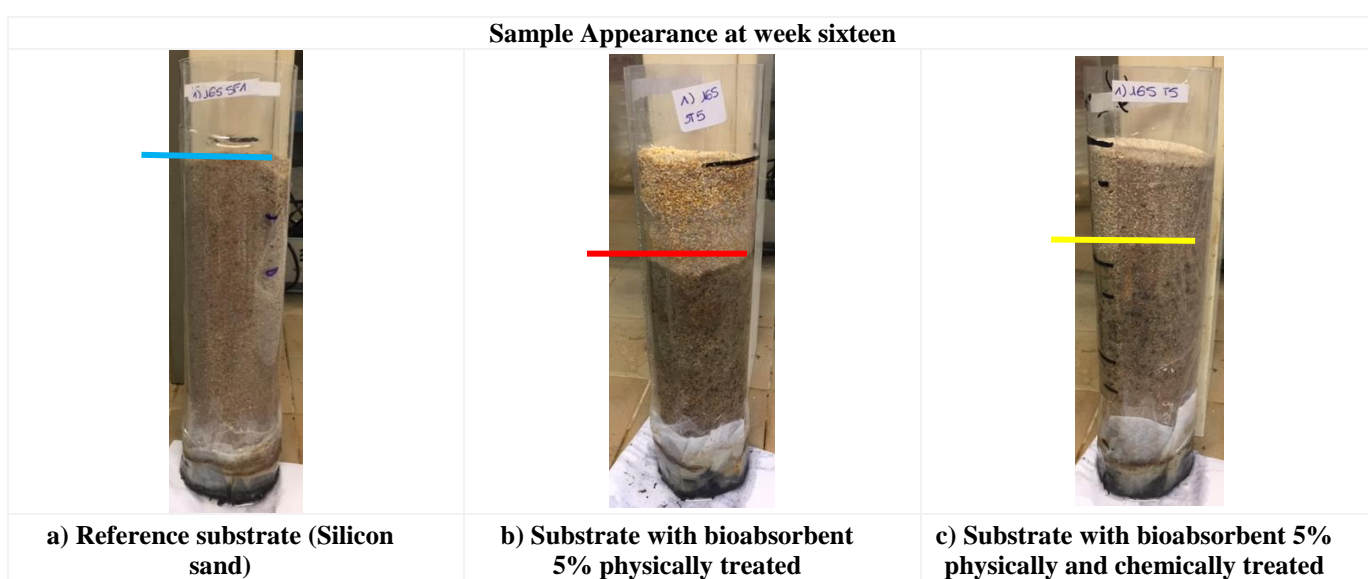


FIGURE 2: Appearance of the samples at week sixteen

The rise of the water corroborated the interaction of the seawater and the bioabsorbent. The parameters measured were the pH, humidity and conductivity. The substrate was inside a PVC tube divided into four sections (Figure 3): the first one between the base of the tube and 11 cm, the second section between 11 – 14 cm, the third between 14 – 17 cm, and the last one on the surface of the tube 17 -20 cm.

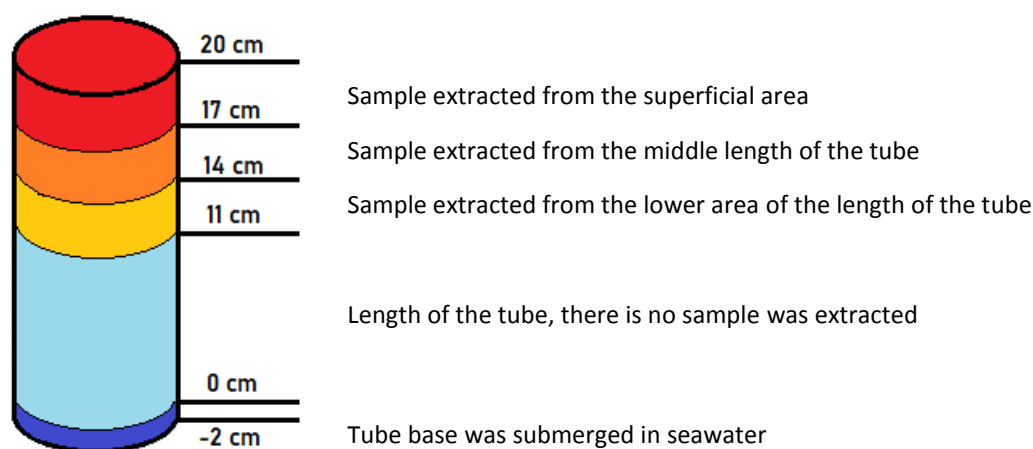


FIGURE 3: Scheme of the samples extracted showing the function of the levels of the tube

The percentage of humidity was determined gravimetrically. The methodology used to do so, was taking a sample with a known weight and gently heating on a stove in the laboratory for 24 hours at 110°C. After this time, the sample was taken through a process of desiccation and then weighed again, with the difference weight showing the amount of water that contained the sample, and so the percentage of humidity. The stove used it during the process was a model Nahita 631/4 and a precision balance, model SCALTEC SBA 52

Once that the sample was dried, the level of salts in the sample could be determined. A known weight of the sample was taken and deionized water was added with a ratio of 1:5 (m/v), then it was agitated for 10 minutes. The next step was to let the silicon sand in the moisture sediment filter the liquid in order to separate the solids in suspension. Both conductivity and pH were measured at environmental temperature using a conductimeter Jeulin JLC20 and a Ph meter Thermo Scientific Orion 2 Star. The values of conductivity could now be used to determine the concentration of salts in the samples with a calibration line.

The pH values of the function of the kind of substrate are shown on Figure 4. There is a significant difference between the pH of the samples without a bioabsorbent, which is nearly neutral and the other samples with bioabsorbent at 5%, whether are chemically or physically treated.

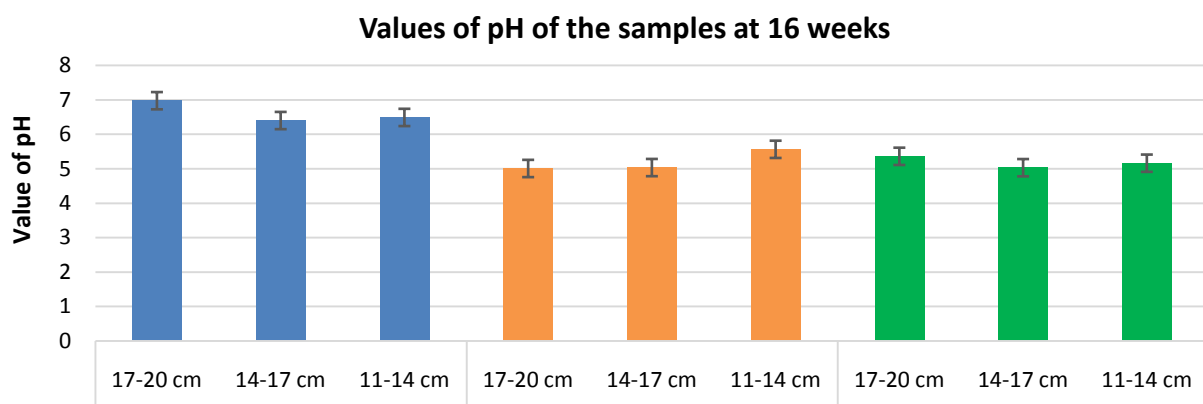


FIGURE 4: Values of pH of the samples at 16 weeks

The pH of seawater is between 7.4 and 8.5, and thus there is a definite interaction between the substrate and the seawater, increasing with the presence of the bioabsorbent. Probably, this interaction might be as a consequence of the microorganisms developed during the sixteen weeks.

Figure 5 shows the results of % humidity and conductivity on the samples extracted at week sixteen. In all the cases, the values of humidity were lower at the surface (17-20 cm), which means that the lower the height of the extracted sample, the higher the humidity.

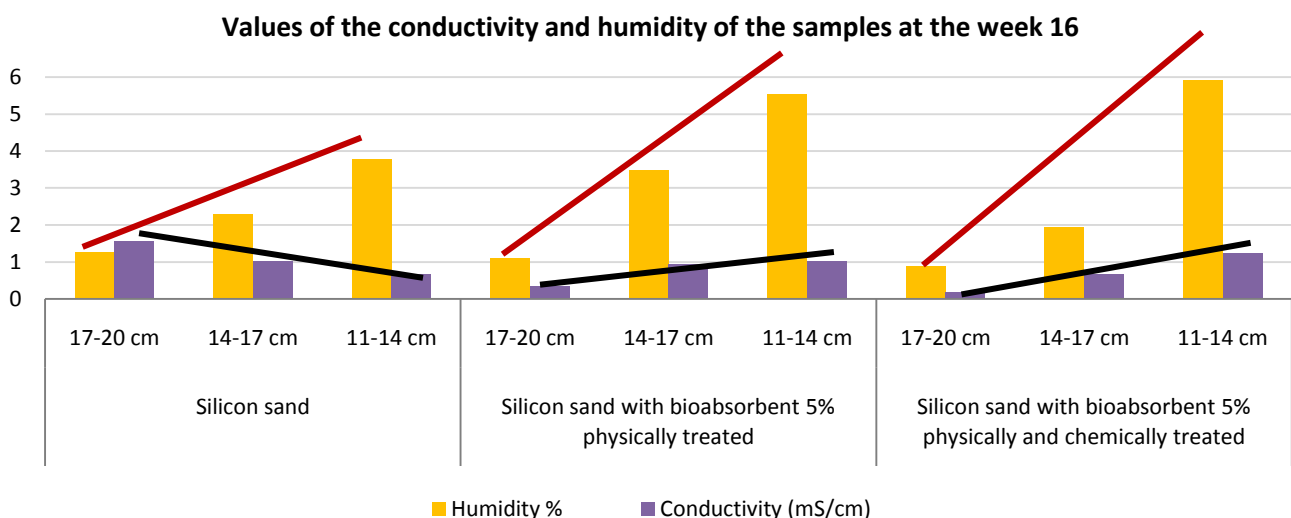


FIGURE 5: Values of the conductivity and humidity of the samples at the week 16

On the other hand, the values of conductivity were to the contrary depending on whether the sample contained bioabsorbent or not in the substrate with just silicon sand, the conductivity increased as did the height of the extracted sample, the highest value was at a level between 17-20 cm. This behaviour was the result of a self-munching effect: the seawater rose to the surface, where it was evaporated and the salts were retained and accumulated, giving an increase in the conductivity value.

The samples with silicon sand mixed with the bioabsorbent at 5% with a physical and/or chemical treatment had the opposite effect to what was seen in the preceding case. The bioabsorbent had an effect in reducing the conductivity as a result of the capacity of absorption which retained salts through the tube (García et al., 2020). It must be noted that seawater has a conductivity of approximately 50 mS/cm. Once it had risen through the tube and reached the surface, the conductivity was below 2 mS / cm, so the reduction in salinity was really important.

3.2 Results at week eighteen, twenty and twenty-four

Furthermore, when the samples were pulled out at week sixteen, 500 mL of deionized water were added to the other samples, imitating rainfall. The soil contained seawater or the salts of it, so the water entered through the pores from the surface level and filtered into an already saturated regimen. When the soil is completely saturated with water from rain or after irrigation, the pores are filled with water and in this condition, as a result of the force of gravity the water descends into lower levels. It was this process that we artificially imitated and observed the effect on the conductivity values.

The aim of the leaching process was to test the capacity of regeneration of the soil, in this case the silicon sand, by measuring the conductivity of the samples. Hence, the conductivity was the most important parameter to be analysed, because it showed if the soil was or was not able to regenerate in situ, because in the case of a big increase in cropping, the irrigation by capillarity would make the soil impossible to be constantly replaced. In Figure 6 the conductivity values of the samples of silicon sand over twenty-four weeks are compared.

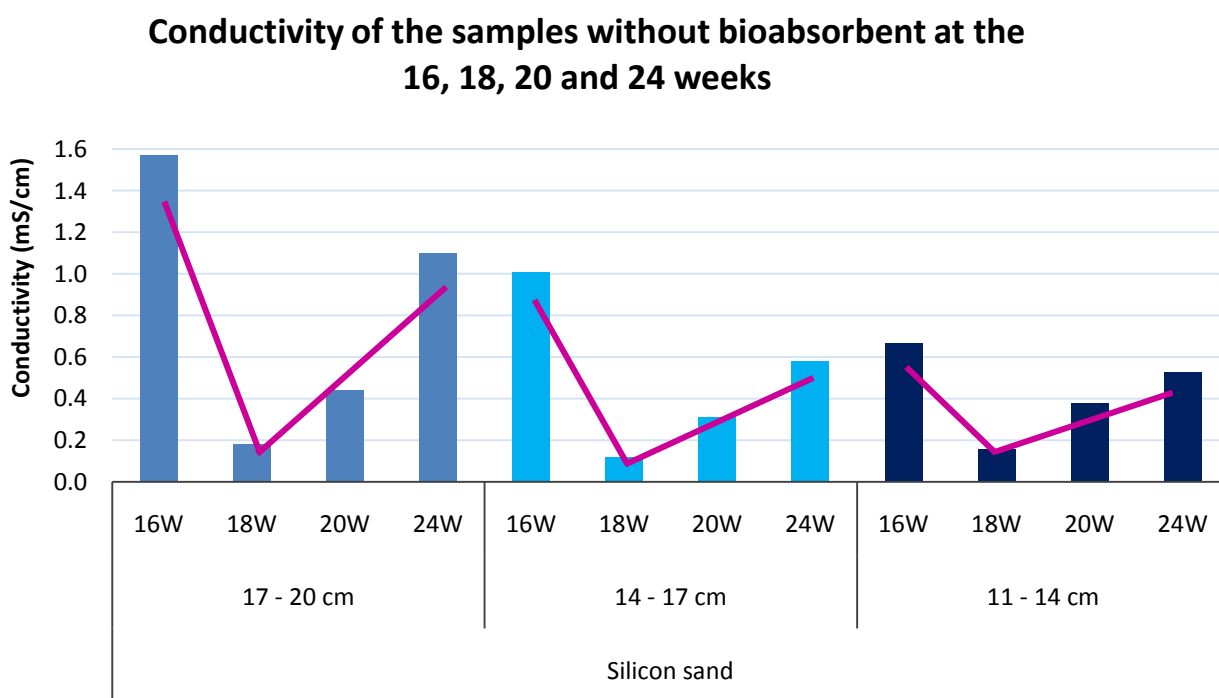


FIGURE 6: Conductivity of the samples without bioabsorbent at 16, 18, 20 and 24 weeks

The conductivity of the sample at week sixteen was the highest in comparison to the samples that were extracted on week eighteen, twenty and twenty-four. The addition of deionized water at that point develops a leaching process, and the minimum value of conductivity was obtained at week eighteen. This parameter increased over the next few weeks. However, the conductivity value at week 24 did not reach that of week 16. Therefore, in those eight weeks after the addition of deionized water on the substrate surface, the conductivity value never reached the experienced value during the first 16 weeks.

The behaviour of the sample without bioabsorbent was the same as the samples that contained bioabsorbent at 5% w/w with a chemical and/or physical treatment (Figure 7).

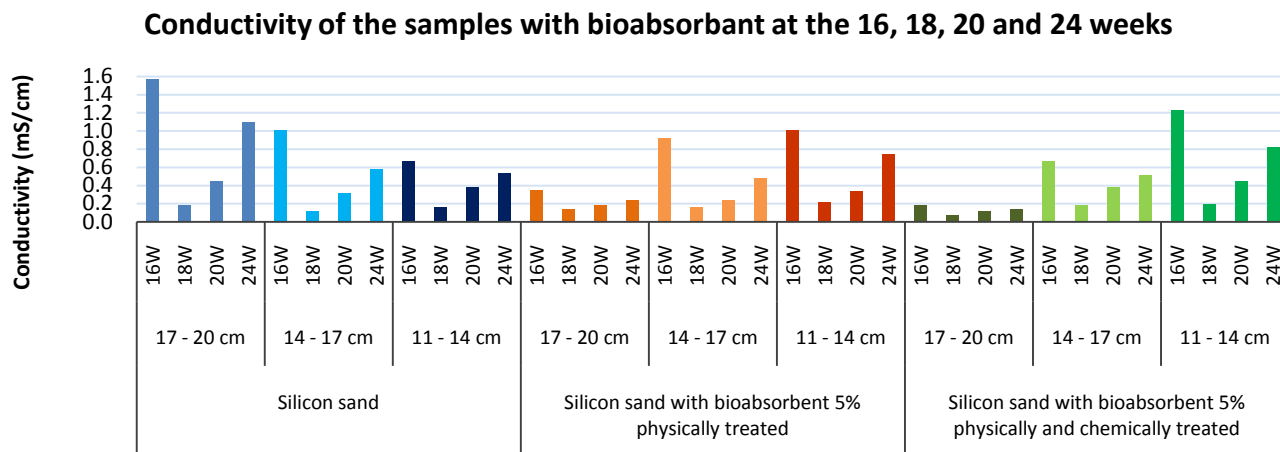


FIGURE 7: Conductivity of the samples with bioabsorbent at the 16, 18, 20 and 24 weeks

Figure 8 shows the pH of the samples studied at the time of the extraction. At week twenty-four the sample without bioabsorbent had stabilized at 7.5. Conversely, the presence of a bioabsorbent that had been chemically and/or physically treated had a significant influence on the values obtained at the base of the PVC tube (11-14 cm). In the middle and higher levels (14-20cm) of the tube, the values were more acidic, which confirmed the interaction of the substrate and seawater as seen before week sixteen, and probably as a consequence of the development of the microorganisms.

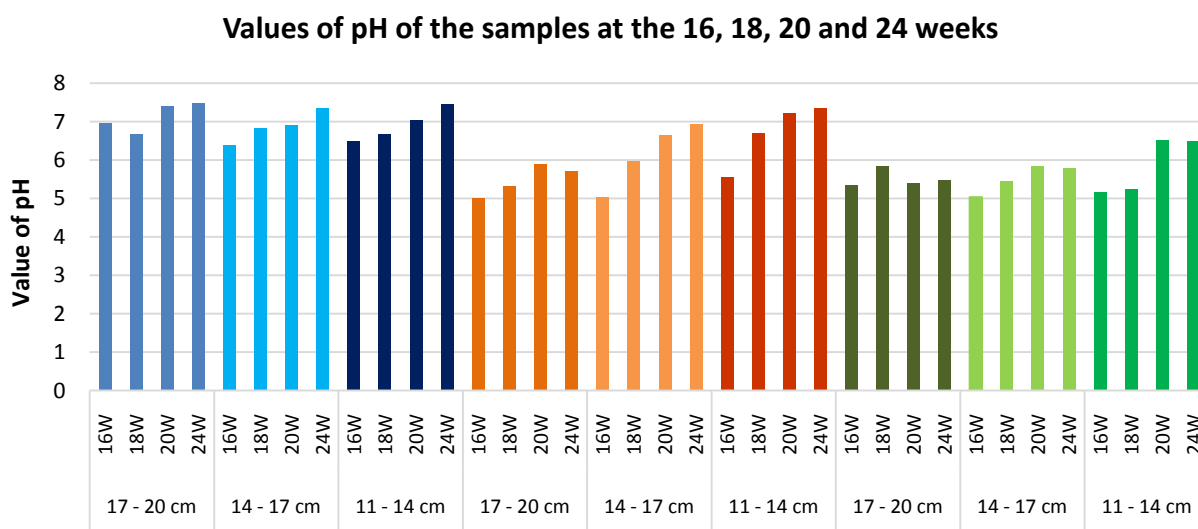


FIGURE 8: Values of pH of the samples at the 16, 18, 20 and 24 weeks

IV. DISCUSSION

Bioabsorption is a physiochemical process like adsorption or ion exchange, which consists of adsorbing chemical into the cellular structure of a biomass like algae, fungi, bacteria, fruit shells, agricultural products or some biopolymers (Chojnacka 2010).

The cellular walls of the bioabsorbents contain polysaccharides, proteins and lipids which have the capacity to bind heavy metals and cationic molecules to the surface. These compounds have functional groups like amines, carboxylic, hydroxyl, phosphates and thiol, each one has a different affinity and capacity to bind different metallic ions (Ghimire 2003).

The orange peel (*Citrus sinensis*) is obtained as a by-product of orange juice production and normally treated as a waste. Even so, from the pectin (citric pectin) can be extracted essential oils, which has a multiple uses in the pharmaceutical and feeding industries. However, orange peels similar to other citric fruits have been used successfully to eliminate heavy metals and textile dyes. (Hameed 2008; Li 2008; Gupta 2009; Lu 2009).

Depending on the source, pectins can vary in molecular size, degrees of acetylation and methylation, galacturonic acid content, and neutral sugar residues. This means that pectin might have versatile gelling properties and form complex compounds with other natural compounds, and therefore, due to their properties, pectins are useful in creating new alimentary products (Gawkowska et al., 2018).

The product obtained after the pectin extraction is a poor food supplement for animals, which has a low protein but high sugar content (Siles et al, 2016). Regardless of that, it has the optimal properties for an after treatment as a bioabsorbent (Masmoudi et al, 2008).

The extraction of the soluble pectin in an acidic medium is necessary to develop its capacity to absorb great quantities of water and form colloids. If it wasn't extracted, the product would not have an appropriate level consistency to be used as a bioabsorbent. The acid hydrolysis makes possible the solubilisation and degradation of carbohydrates, especially xylan and hemicellulose, since glucomannan is a relatively stable compound in an acidic medium. (Van Buren, 1991).

An initial acidic treatment is required to be completed in order to increase the absorption capacity of the citric peel, and then the next phase is an alkaline treatment. The aim of the process in an alkaline medium is to saponify the functional groups of esters, which have not been extracted during the original acidic process (Cardona Gutiérrez et al, 2013). In general, the NaOH is used for the alkali process, added after the CaCl₂ with the objective of increasing the activation. However, it has been already demonstrated that both process might be done in a combined phase using the Ca(OH)₂ 0.2 M (Arjona et al., 2018).

The use of a bioabsorbent 5% w/w with the silicon sand improved the behaviour of the substrate decreasing the salinity of seawater. The results of the tests of the bioabsorbent with physical treatment (orange peel subjected to a drying process to give a constant weight plus subsequent grinding with a granulometry between 500-1000 µm), were improved using a bioabsorbent with a chemical treatment and a granulometry between 500-1000 µm too.

In all the cases, the experimental values of conductivity were under 2 mS/cm (Table 1), this is the limit value for successful cropping without negative consequences for the plants, and it is important to note that a lot of water for drainage, including subterranean sources is between 2-10 mS/cm (Rhoades et al., 1992). Obviously, to successfully develop this new technique of irrigation, the substrate will have to be optimized in order to use it on an industrial scale and also have the water and nutrients available which are necessary for successful cropping. The tolerance salinity of the soil will have to be revised so as to not exceed the limits of the values for successful cropping.

TABLE 1
TYPE OF SOIL IN FUNCTION OF CONDUCTIVITY

Soil type	Salinity	Conductivity (mS/cm)	Effects	Crops
Normal	Very light	0-2	No effects	Corn, lemon, apple, peach ...
	Light	2-4	Might affect sensible crops	Rice, olive trees, tomato plants, melon, spinach ...
Saline	Medium	4-8	Effects majority of crops	Wheat, soy, cotton, barley, beet ...
	Strong	8-16	Cropping is difficult	-----

V. CONCLUSIONS

- The use of seawater for capillary irrigation is possible as is demonstrated by this project.
- The soil must have specific characteristics in composition and granulometry in order to reduce the salinity. At the same time, the soil must keep the humidity at a certain level for the roots of the crops, because of the need for hydration and nutrient absorbance.
- If the seawater does not show a decrease in salinity it would be completely detrimental to crop growth. Silicon sand meets these requirements and can be regenerated by leaching with fresh water. In addition, this kind of soil or filter medium, silicon sand, can be found all over the world and is an economic and abundant medium.
- From a height of 10 cm from the seawater level, the experimental values of electrical conductivity were less than 2 mS/cm.
- The mix of the silicon sand and bioabsorbent obtained from orange peel contributes efficiently in the retention of salts from the seawater. This type of filter medium contains two components that are easy to find: silicon sand, which can be found all over the world, and orange peel is a poorly used waste from the food industry.

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Production of Vermicompost from Rose Flower Petal Wastes

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Abstract— The flower waste obtained from markets and temples could be effectively utilized for vermicomposting and production of good quality vermicompost. *Eudrilus eugeniae* species is good for vermicomposting of flower waste at shorter time period. The results obtained proved that 30% inclusion of flower waste along with cow dung gives good yield of vermicompost. *Eudrilus eugeniae* does not require soil for habitation. Provision of good quality cow dung enhances the water holding and nutrient supplying capacity. Maximum temperature (27.96°C) was recorded in VT6 and pH ranged between 7.58 and 8.76 in all treatments. VT6 treatment also showed the maximum electrical conductivity (3.94 mhos/cm). 42.50% of Organic Carbon was observed in VT2. A high concentration of N (0.59%) was found in VT5 treatment and phosphorous (0.68%) in VT4. The maximum weight of earthworm (3080 mg) was achieved in VT5 with a growth rate of 24 mg/worm/day. High yield of vermicompost (1422gm/2kg of substrate) was also obtained in VT5 treatment. The present study revealed that the temperature at a range of 26°C, pH 7.5 to 8 and moisture content of 49 to 50% were the ideal parameters to activate metabolic activity, cocoon production and reproductive action of *Eudrillus eugeniae*..

Keywords— Earthworm, *Eudrillus*, Vermicomposting, Flower wastes, *Rosa berberia*.

I. INTRODUCTION

Floriculture is an art and science and it has been recognized as an economic activity with the potential for creating employment. In Tamil Nadu flowers like Marigold, Tuber Rose, Jasmine, Mullai, Rose, Crossandra, Chrysanthemum, Arali, etc are cultivated in large numbers. Tamil Nadu takes third place by cultivating the flowers in an area of 32,290 ha. Flowers are marketed regularly in local market and mainly operated in open yards like road sides. The price of flowers mainly depends on the quality, supply, demand and freshness. Heavy rainy season and too hot summer season results in deterrent for selling flowers. Floral waste generation also occurs largely during functions, worships, ceremonies, festivals, etc. This ultimately results in the large amount of flower waste which creates severe environmental pollution and health hazard. Biological processes such as vermicomposting converts floral waste into organic fertilizer would be of great benefit.

Vermicomposting is a biological conversion process of organic materials and involves a joint action of earthworm species and microbes, which is being used for solid waste management [1]. Floral waste degradation is a slow process as compared to kitchen waste degradation [2]. Therefore there is a need of proper and eco-friendly method for floral waste degradation. In this the present investigation is aimed to document the stepwise physico chemical changes during floral waste degradation and vermicompost production by using an indigenous species of earthworm

II. MATERIAL AND METHODS

2.1 Collection of rose flower waste

Rose (*Rosa berberia*) flower waste was collected from local flower market and temples at Salem. About 10kg was collected and the non-biodegradable part of rose flower waste was removed by hand sorting and the biodegradable waste were segregated and shredded into small pieces.

2.2 Collection of earthworms

Earthworm species *Eudrilus eugeniae* species was procured from local suppliers at Salem, Tamil Nadu, India.

2.3 Combinations of composting substrate

For vermi-composting, rose flower waste and cow dung was mixed in different ratios. These rose floral waste substrates were filled in the plastic bins.

- VT 1 – Positive control (Cow dung alone + Earthworm)
- VT 2 – Negative control (Rose flower waste alone + Earthworm)
- VT 3 - 10% Flower waste (200g Flower waste +2kg Cow dung + Earthworm)
- VT 4 - 20% Flower waste (400g Flower waste +2kg Cow dung + Earthworm)
- VT 5 - 30% Flower waste (600g Flower waste +2kg Cow dung + Earthworm)
- VT 6 - 40% Flower waste (800g Flower waste +2kg Cow dung + Earthworm)
- VT 7 - 50% Flower waste (1000g Flower waste +2kg Cow dung + Earthworm)

Each combination was prepared in triplicates. The bins with different combinations were left for 12 days prior to experimentation and watering was done on alternate days. Then they were mixed upside down for pre composting, microbial degradation, softening of waste and for thermo stabilization

2.4 Preparation of composting substrate

The experiment was performed in plastic containers (60cm length x 30cm width x 30cm depth) with uncovered top surface. The vermibed substrates (VT1 to VT 7) consisted of rose flower waste and cow dung in different combinations by dry weight as described above. These wastes were used to provide bedding for the earthworms as well as a carbon supplement. Cow dung was used as inoculants. Alternate two layers of floral waste and cow dung are placed one over another. Adult clitellate worms, *Eudrilus eugeniae* ranging in length from 12cm to 16.7cm were added at the rate of 100 grams of earth worms per 1Kg of composting substrate. The moisture content was maintained throughout the period of the study by periodic sprinkling of water. Watering was stopped, when the vermicompost got ready as indicated by uniform dark brown to black colored granular structure. The vermin cast was passed through 2-3 mm sieve and the earthworms were removed manually. The vermin cast was air dried by spreading it in large trays. The ratios of bioconversion rate of flower waste into vermicompost for all the groups were calculated. After sufficient moisture was lost, samples were analyzed. Cow dung substrate alone was used as positive control. Rose flower waste substrate alone was used as negative control.

2.5 Physiochemical parameters

Gravimetric method was used for determining the moisture content. Temperature was measured with the help of Mercury thermometer at the depth of 10 cm and their mean values were taken in centigrade [3]. Vermicompost were oven dried at 100°C then ground in the blender and sieved for its physiochemical analysis. Particles smaller than 2mm in diameter were used for analysis. Electrical conductivity was determined by using pH and electrical conductivity meter. 10 grams of dried sieved vermicast sample was diluted to 100 mL with distilled water (1:10 w/v) and kept for shaking in a rotary shaker for two hours. After two hours the sample was left for half an hour for settling and then filtered through a whatman filter paper no 42. The filtrate was used for pH and EC analysis [4]. Total organic carbon (TOC) was measured by the method of Nelson and Sommers [5]. The total nitrogen of the sample was estimated by Kjeldahl method. Total potassium content of the was determined by flame photometric method and total phosphorous content was estimated by diacid digestion method.

2.6 Growth performance and Cocoon production rate

The growth and reproductive potential of earthworms in different treatments were determined. At the end of the experiment, adult worms and cocoons were counted separately. The worms were separated from the vermicompost and earthworm biomass was determined. The substrate in treatments was examined daily in order to determine the onset of cocoon production. When the cocoons appeared, they were picked up by hand sorting, washed lightly in distilled water and counted so as to determine the total number of cocoon production rate (cocoon/worm/ day). At the end of the experimental day, the earthworms were collected from each treatment bins and rinsed with distilled water to remove any adhering substances, dried briefly on paper towel and weighed using electronic balance. Without voiding their gut content the worms were weighed. Weighed earthworms were again introduced in each respective experimental container. From the data collected, the increased biomass rate (g/ earthworm/day), weight achieved at the maximum, and net biomass gain were calculated.

Growth rate determination, $R = (N_2 - N_1) / T$

Where R = Growth rate; N1= Initial biomass of earthworm (mg); N2= Final biomass of earthworm (mg); T= Time period of the experimental day

III. RESULTS

Experiments were conducted in plastic containers using different combinations of rose flower waste along with cow dung. Triplicates were taken for each test treatment. Cow dung alone was used as positive control and rose flower waste alone was used as negative control. After a pre-decomposing period of 15 days, 100 grams of clitellate earth worms were released in to bin per kilogram of composting substrate. During this period no mortality was observed, earthworms could survive in all treatments except negative control. Rose flower waste alone was not found suitable and all the earth worms did not survive.

3.1 Physio- chemical characteristics of vermicompost

The variation of pH, EC, Temperature and Moisture content in different substrate material (flower waste & cow dung) including controls are presented in table-1. The pH values ranged between 7.58 and 8.76. The maximum pH 8.76 was recorded in control treatment (VT2) and the minimum pH of 7.66 was noticed in VT7 treatment. The pH value gradually decreased from 8.76 to 7.66 during the end of the experiment days. The pH shifted towards acidic range which might be attributed to mineralization of the nitrogen and phosphorus into nitrites/nitrates and orthophosphates. pH in the final vermicompost samples might have been changed due to the microbial activity during the bioconversion of different substrates.

TABLE 1
PHYSICAL COMPOSITION OF VERMICOMPOST SAMPLES IN DIFFERENT TREATMENTS

S.No.	Compositing Substrate	pH	EC (mhos/cm)	Temperature (°C)	Moisture (%)
1	VT 1	00	00	00	00
2	VT 2	8.76±0.32	1.52±0.02	27.29±0.50	40.33±1.52
3	VT 3	8.37±0.12	2.64±0.23	25.97±0.15	42.33±0.57
4	VT 4	8.00±0.11	2.19±0.03	25.67±0.58	44.33±2.52
5	VT 5	7.58±0.20	2.95±0.13	26.20±0.86	49.66±1.52
6	VT 6	7.78±0.22	3.94±0.06	27.96±0.06	50.33±1.52
7	VT 7	7.66±0.12	3.76±0.12	27.50±0.50	50.33±1.52

The electrical conductivity (EC) of vermicompost ranged between 1.52 and 3.94 mhos/cm in various treatments. The maximum electrical conductivity (EC) 3.94 mhos/cm was obtained in VT6 treatment and the minimum electrical conductivity (1.52) were found in VT2.

The temperature variations indicated that it was higher in larger proportions of rose flower waste and it decreased as the amount of cow dung increases in composting mixture. The highest temperature of 27.96°C was noted in VT6 and the lowest temperature (25.67°C) was found in VT4 treatment at 45th day of experiment. Another significant observation was that the temperature increased (27.9°C) in earlier period of composting and then decreased to 26.2°C in later period of Vermicomposting.

The moisture content of the wastes varied between 40.33% and 50.33%. The highest moisture content (50.33%) was recorded in VT6 & VT7 treatments whereas lowest moisture content (40.33%) was found in negative control treatment. The present study high moisture (50%) content in VT6 than the control, which may be due to the assimilation rate by microbial population indicating the higher rate of degradation of flower waste by earthworm.

The NPK Contents of the harvested vermicompost was depicted in table-2. This shows a significant difference between the treatments. The concentration of N was highest (0.59%) in VT5 treatment. On the other hand, the amount of in P and K were high (0.68 and 1.62%) in VT4 & VT3 treatments and low Nitrogen (0.39%) in VT7 treatment. The maximum percentage (42.50%) of Organic Carbon was observed in VT2 treatment and minimum percentage (29.16%) of organic carbon was observed in VT7 treatment.

TABLE 2
CHEMICAL CHARACTERISTICS OF VERMICOMPOST SAMPLES IN DIFFERENT TREATMENT

S. No.	Compositing Substrate	Total Organic Carbon (%)	Total Nitrogen (%)	Total Phosphorus (%)	Total Potassium (%)
1	VT 1	00	00	00	00
2	VT 2	42.50±0.50	0.43±0.01	0.44±0.05	0.91±0.03
3	VT 3	41.76±0.68	0.55±0.04	0.41±0.01	1.62±0.14
4	VT 4	39.67±0.58	0.53±0.03	0.68±0.03	1.17±0.11
5	VT 5	39.33±1.15	0.59±0.02	0.60±0.04	1.60±0.52
6	VT 6	31.00±1.00	0.46±0.04	0.56±0.05	0.67±0.04
7	VT 7	29.16±1.04	0.39±0.01	0.45±0.05	0.76±0.16

3.2 Yield of vermicompost

Vermicompost production rate was different according to the composition of flower waste mixture (Figure-1). The highest vermicompost yield (1422 gms) was obtained from VT5 treatment followed by 1213 & 1100 gms were harvested in VT4 & VT3 respectively. The lowest vermicompost yield of 809 gm was noticed in VT7 treatment.

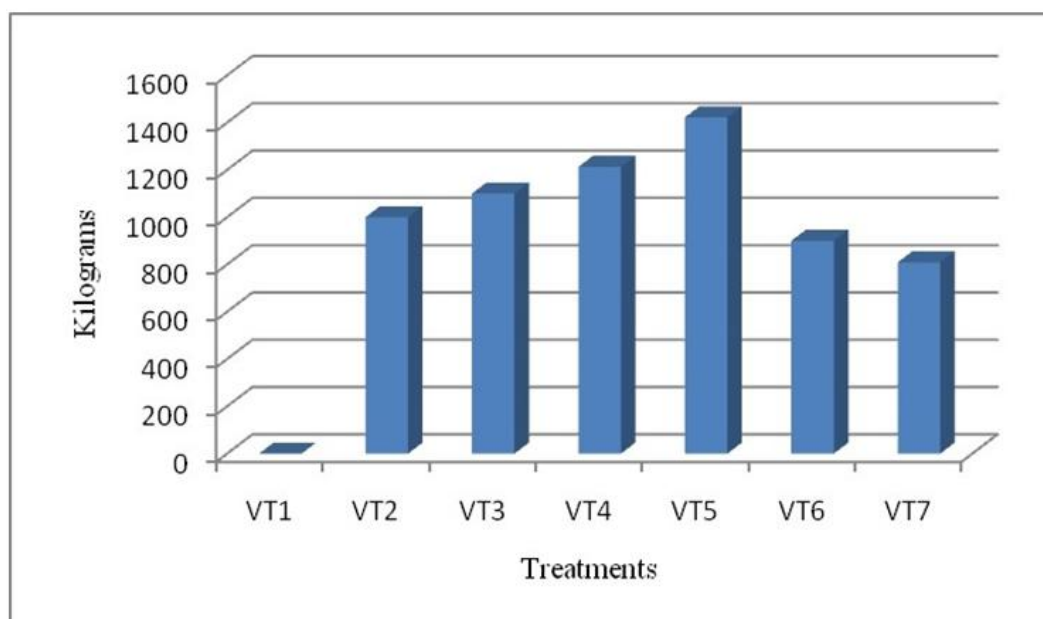


FIGURE 1: Yield of Vermicompost in different combination of Flower Waste Substrate

3.3 Growth and reproduction rate of *Eudrilus eugeniae*

The growth rate (mg weight gained/day/earthworm) has been referred as a good comparative index to study the growth of earthworms in different waste substrates. The growth rate of earthworm species in different substrates during the study period is given in Table-3. The biomass of earthworm species showed progressive increase to 3083mg on 45th day of experiment with a growth rate of 24 mg/worm/day in VT5. The minimum weight was observed as 2325 mg of earthworm with the growth rate of 7.22 mg/worm/day in VT2. These values showed a gradual decline (10.22mg/worm/day and 12.00mg/worm/day) with increasing proportion (40 & 50%) of rose flower waste. However, in VT2 the weights (7.22mg/worm/day) of adults were less when compared to treatments. The increased biomass with 30% flower waste suggests that the role of flower waste as food material and enriching the substrate with essential nutrients.

TABLE 3
BIOMASS GAIN OF *EUDRILUS EUGENIAE* IN DIFFERENT TREATMENTS

S. No.	Compositing Substrate	Initial Biomass of Earthworm (mg)	Final Biomass of Earthworm (mg)	Net Biomass gain of Earthworm (mg)	Biomass increase rate (gm/Earthworm/day)
1	VT 1	2000	00	00	00
2	VT 2	2000	2325.00±25.00	325.00±0.05	07.22±0.55
3	VT 3	2000	2476.60±58.60	476.66±58.59	10.59±1.29
4	VT 4	2000	2818.00±38.31	818.00±38.31	18.18±0.85
5	VT 5	2000	3080.00±29.00	1080.66±29.00	24.00±0.65
6	VT 6	2000	2542.00±74.84	542.66±74.84	12.00±1.67
7	VT 7	2000	2466.00±76.60	466.00±76.60	10.22±1.76

Table 4 shows the production of cocoons. High yield of cocoon production (374 numbers) was attained in VT4 treatment at a reproduction rate of 3.45 (cocoon/earthworm/day) at 45 day of experiment. The lowest number (92 numbers) of cocoon was found in VT7 treatment with a reproduction rate of 1.17 ± 0.17 (cocoon/earthworm/day). The mean number of cocoons produced per worm per day was 12.08 ± 0.32 in VT4 treatment which was greater than 2.96 of cocoons produced per day in VT7.

TABLE 4
REPRODUCTION RATE AND COCOON PRODUCTION OF *EUDRILUS EUGENIAE* IN DIFFERENT TREATMENTS

S. No.	Compositing Substrate	Cocoon Production started (week)	Total Number of Cocoon Produced (Numbers)	Number of Cocoons produced/day	Reproduction rate (Cocoons/worm)
1	VT 1	00	00	00	00
2	VT 2	2	121.00±06.55	03.89±0.21	0.46±0.05
3	VT 3	2	333.00±11.35	10.74±0.36	1.32±0.40
4	VT 4	2	374.00±10.00	12.08±0.32	3.45±0.38
5	VT 5	2	342.00±29.00	11.05±0.95	2.37±0.55
6	VT 6	2	143.00±13.52	04.61±0.43	1.89±0.11
7	VT 7	2	92.00±17.43	02.96±0.56	1.15±0.17

IV. DISCUSSION

Vermicompost is considered to be the high value product and is an excellent soil additive produced by earthworms. The present deals with the physico chemical changes during floral waste degradation and vermicompost production by using an indigenous species of earthworm.

4.1 Physico-chemical parameters

In the present investigation pH values were ranged between 7.58 and 8.76 in all treatment groups. The results of the present investigation corroborates with the findings of Vodounnon *et al.*, [6] who reported that the pH values of organic substrate and vermicompost ranged between 7.94 to 8.52 which was due to higher mineralization. vermicomposts tend to have pH values near neutral, which may be due to the production of CO₂ and the organic acids during microbial metabolism [7]. The electrical conductivity (EC) of vermicompost ranged between 1.52 and 3.94 mhos/cm in different treatments. The increased

electrical conductivity (EC) during the vermicomposting process is in consistence with that of earlier studies [8],[9]. In this study our findings correlates with previous work of Sharma and Yadav [10], who has reported that the electrical conductivity of cow dung vermicompost was 5.44 and 4.87 mS/cm, respectively. The main reason for the increase in EC might have been due to release of different mineral salts in available forms [11]. Their concentration increased gradually with time, which may contribute the increase in electrical conductivity of compost.

The temperature in the present study ranged between 25.6°C to 27.9 °C temperature plays a vital role in vermicomposting. The increase in temperature can kill the earthworm. Similar types of results were observed from temple waste [12]. Nisha Jain [13], has also reported that 25°C was found to be optimal for flower waste composting substrate material. Similar changes were also observed in temperature during vermicomposting, they reported that initial temperature (30°C) of the vermibed was found to decrease up to 26°C because of exothermic process of organic matters [14]. Relatively higher moisture content of vermicompost produced by *Eudrilus eugeniae* implied greater palatability of the substrate (VT5 treatment). The optimum moisture content for growth of earthworm *Eudrilus eugeniae* was 85% in organic management [15]. Vermicompost samples of the present study showed higher (50.33%) moisture content, which may be because of assimilation rate by microbial population indicating the higher rate of degradation of flower waste by earthworm.

The NPK Contents (%) of the harvested vermicompost showed a significant difference between the treatments. The concentration of N was high (0.59 %) in VT5. In a study by Manaig [16], reported that the NPK content of vermicompost is higher than the farmyard wastes. Sharma and Yadav [17], also reported an increased total nitrogen content in vermicomposts. Earthworms in waste material considerably enhances the amount of N due to earthworm mediated nitrogen mineralization of wastes and also the earthworm enhances the nitrogen levels of the substrate by adding their excretory products [18]. Total nitrogen content in vermicomposts can range quite widely from 0.1% to 4% or more [19]. High K content (0.68%) was recorded might be due to the high microbial activity which favours mineralization. Chaulagain *et al.*, [20] reported that there is increased potassium content of the vermicompost using cow dung with banana pseudostem than other wastes. There is an increased phosphorous content in vermicompost also recorded in the present study. The increased phosphorous might be probably due to mineralization and mobilization of phosphorus as a result of bacterial and faecal phosphatase activity of earthworms [21]

4.2 Growth Rate, Biomass and Cocoon Production

The increased biomass of *Eudrilus eugeniae* (3080mg) was observed in VT5. The growth rate and the biomass of earthworm species during the study period showed progressive increase up to 4th week. Similar pattern of results were also reported [22], [23] and suggested that the rapid pre-reproductive phase of growth is followed by a phase of progressive biomass and growth reduction once sexual maturity was attained. The loss of biomass might be associated with the exhaustion of food. Neuhauser *et al.*, [24] reported that the weight reduction also occurred when the earthworms attained their matured stage and also due to utilization of energy for the purpose of reproduction such as laying eggs, mating and cocoon formation [25]. Temperature also influences the growth of earthworm by modifying metabolic activity [26]. Vasanthi *et al.*, [27] also reported that the earthworm growth ability was high at 26°C. So temperature also plays a vital role in the growth rate and biomass of the worms.

At the end of experiment (45 days) a high number of cocoon (374 numbers) productions were obtained in VT4 with a reproduction rate of 3.45 ± 0.38 (cocoon/earthworm/day). A high number of worms and cocoons were observed in cow dung and saw dust mixture (1:1 and 1:1.5) than the pure cow dung using epigeic earthworm *Eudrilus eugeniae*[28]. Also, high numbers of earthworms were observed in the feeding mixture of cow dung and leaf litter as compared to the cow dung as a sole feeding material. The presence of leaf litter in the mixture which may be favored earthworm multiplication. In a similar study, the maximum increase in population of *Eudrilus foetida* was observed in tree leaves as a sole feeding material than cow dung alone [29]. Suthar [18] reported that the nitrogen content of substrates is an important factor related to cocoon production. Getachew *et al.*, [30] have reported that food source also greatly influences growth rate and cocoon production. Survival, biomass and reproduction of earthworms are the best sign to analyse the vermicomposting process. Therefore, the increase in adult worm biomass and the production of significant numbers of cocoons in VT5 indicates that their reproductive activities are enhanced by the substrate. Our present study is also supported by the findings of Loni *et al.*, [31] who have reported that the total number of cocoons produced were lower in higher pH (alkaline) when compared to neutral pH (6.5 to 7.0). The present study revealed that the yield of vermicompost was higher in 30% flower waste and cow dung combination. So the ability of worm to consume and convert the wastes into vermicast varies according to the substrate and hence the difference in vermicompost yields.

V. CONCLUSION

The present study revealed that the flower wastes may be used as potential substrate for vermicomposting. Vermicompost also contains some of the secretions of worm and in association with microbes acts as growth promoters along with other nutrients. The physico chemical parameters such as pH, EC, temperature and moisture content were tested and found that the *Eudrilus eugeniae* were grown well in the pH of 7.58 ± 0.20 , 2.95 ± 0.13 moh of EC, $26.20 \pm 0.86^{\circ}\text{C}$ of temperature and 49.66 ± 1.52 % of moisture content. Good yields (1322gm/2kg of substrate) of vermicompost were obtained in VT5 treatment (30% rose flower waste inclusion). This ratio is recommended for vermicompost generating profits. The present study divulge that the temperature at the range of 26 to 27°C , pH 7.5 to 8.0 and moisture content 49 - 50% were the ideal parameters to activate metabolic activity, cocoon production and reproductive action in *Eudrillus eugeniae*.

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Striving for restoration of wetlands functions and values in the City of Kigali

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Abstract— *This work aimed to present the commitment of the Government of Rwanda of relocating all illegal activities from wetlands in the framework of environmental protection; to highlights human activities established into wetlands of Kigali City and their categories; and to mention some initiatives of restoration. Its scope is limited to governmental policies presentation and analysis, to the presentation of different activities that degrade wetlands in the City of Kigali and to highlight some initiatives for restoration.*

Documentation, camera and field survey were used in data collection and ArcGIS 10.2 was used as software for spatial analysis and presentation and the survey covered all districts of the City of Kigali. Activities that harm wetlands include: residential home, commercial activities, industrial activities, parking, garages, ware houses, carpentry and welding workshops, dumping sites, bricks burning, petrol stations, carwash, schools, health centers, worship houses, domestic animal growing and play grounds.

Relocation of these activities is the enforcement of the environmental law and the government commitment to environment management. Some of these activities were legally established and their relocation has to be compensated and those illegally established will not be compensated. Many of these wetlands will be left into conservation in order to recover their functions of storing and releasing water and buffering the impacts of floods; providing habitat for plants and animals; providing water storage, improve water quality and reduce pollution, etc. Some few other will be made up into recreational areas like Nyandungu Recreational Park, Kimicanga Entertainment Center and Gikondo wetland parks.

Keywords— *biodiversity, ecosystem, Kigali, restoration, wetlands.*

I. INTRODUCTION

Wetlands are aquatic ecosystems with plants, animals and soils that are adapted to wet conditions which often require and can survive permanent or periodic inundation. Water in wetlands can be still or flowing; it can be fresh, salty or brackish. Wetlands do not have to be continuously wet; some wetlands can remain dry for years at a time [1].

Wetland functions and values are the roles that a wetland performs resulting from specific characteristics of the wetland and the wetland's watershed. Functions are self-sustaining properties of a wetland ecosystem that exist in the absence of society without regard to subjective human values. Values are the worth, merit, quality, or importance of a wetland to society based on either one or more functions and physical characteristics associated with the wetland [2].

Wetland ecosystems represent 4% of Earth's surface, yet comprise approximately 45% of the realized value of natural ecosystems. Wetlands provide important functions such as filtering contaminants, removing nutrients and sediment from runoff, contributing to groundwater recharge, storing floodwater, stabilizing shorelines, and providing habitat for numerous fish and wildlife species [3].

In particular, urban wetlands provide a variety of benefits and services to the community. In addition to providing habitat for plants and animals, wetlands provide water storage, improve water quality and reduce pollution. Wetlands also protect against natural hazards, slowing floodwaters, reducing the risk of fire and protecting against erosion of river banks and coastlines. Wetlands and associated vegetation can provide a cooling effect to surrounding areas in summer and also moderate strong winds.

Wetlands also contribute to the well-being of the community by acting as urban green spaces which provide aesthetic appeal, landscape diversity and recreational opportunities. They also contribute to cultural heritage, spiritual values and they provide easily accessible educational opportunities to learn about the environment [4].

As a beautiful place to live and work in, over 13% (8,740ha) of the City of Kigali is covered by wetlands [5]. Kigali has contributes unique attractiveness of landscapes. The City of Kigali; the Capital of Rwanda is built on hilly area, sprawling across about four ridges and the valleys in between. These Wetlands are Kigali's key hydrological features. These wetlands have important environmental functions, such as storing and releasing water and buffering the impacts of floods. They have been threatened by human activities including the conversion to agriculture, human settlements and industrial uses, and when used for livestock activities and sand quarries. As a result of such impacts, by 2006, only 24 per cent of Kigali's original wetland areas remained [5].

For instance, the urbanized sections of the River Nyabugogo system have been found to be high in heavy metals, such as lead and chromium, and nutrients from organic material and soil runoff [6]. Polluted water exposes households to the risk of contracting diarrheal and other water-borne diseases, especially since 30.2 per cent drink this water without any prior treatment (NISR, 2012a). The prevalence of diarrheal disease in children in urban areas is 9.8 per cent [7].

This work aims to present the will and commitment of the government of Rwanda for relocating all unnecessary activities from wetlands across the country and in Kigali particularly in the framework of environmental protection especially for conservation of the existing swamps; to highlights different human activities established into wetlands of Kigali City and their categories; and to mention some initiatives of restoration that have started.

The scope of this work is limited to governmental policies presentation and analysis, to the presentation of different activities that pollute or destroy wetlands in the City of Kigali and their categories and to highlight some initiatives for restoration. The analysis will be based on the survey outcomes; the mentioned survey has been conducted in July – August 2017.

II. MATERIALS AND METHODS

The survey covered all wetlands in the three districts of the City of Kigali: Nyarugenge, Kicukiro and Gasabo; and it took 35 days with 3 teams from different public institutions. Data collected on every activity were concerning, in addition of location, activity owner, year of establishment, material in which the stuff is made, the document related, number of the parcel, etc.

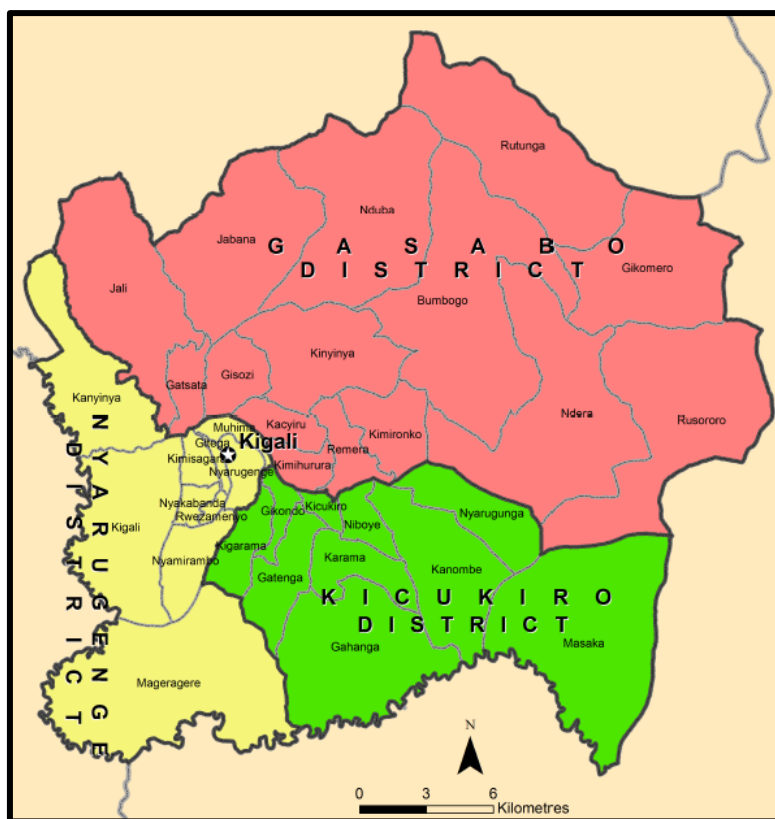


FIGURE 1: Administrative Districts and Sectors of Kigali

In addition of the form for identification, in this work we used the Kigali Master Plan in order to identify limits of wetlands and the planned land use for inventoried activities. We also used cameras for taking some photos that will illustrate what we saw. The ArcGIS 10.1 software has been used for producing some illustrative maps of the work.

Field survey has followed these steps: Physical checking on swamps to highlight the unlawful activities conflicting with the map of swamps, record them and propose a solution in order to relocate them.

III. RESULTS

The conducted data collection has shown that at least 13.8% of the surface area of the City is covered by swamps. The following table indicates the statistics for swamps situation in each District of the City of Kigali. The big number of parcels is to be found in the swamps buffer because it is easy to subdivide land in swamp buffer than the one completely falls within the swamp. The other reason behind should be the way an area was occupied before the systematic land registration of 2010.

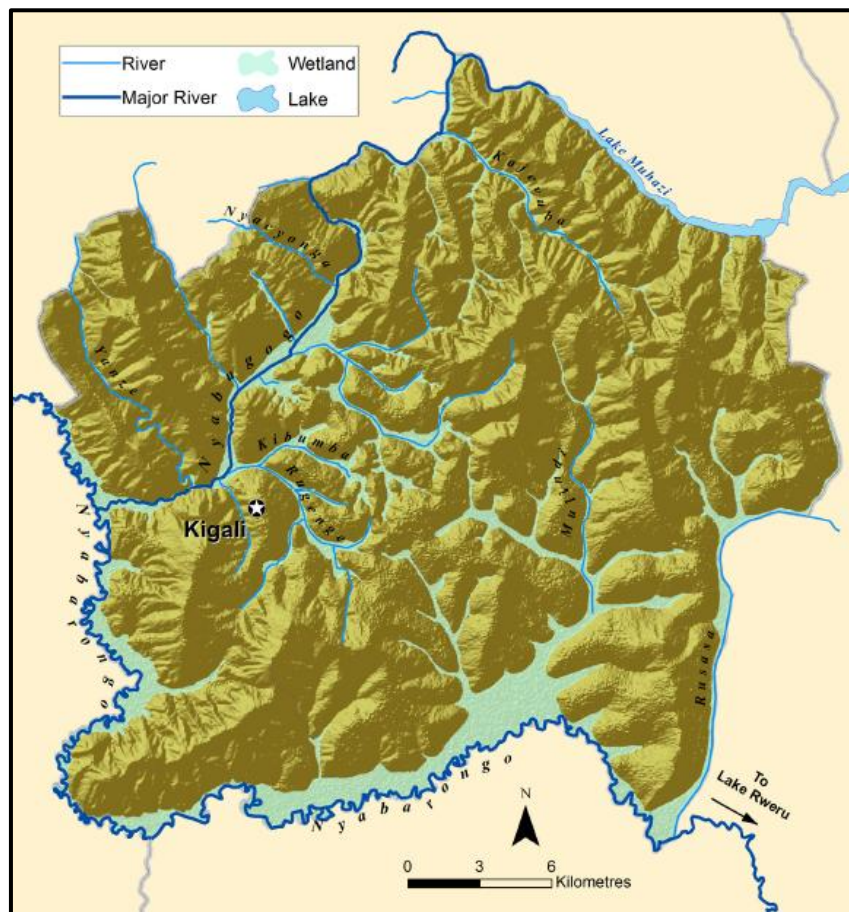


FIGURE 2: Kigali's network of rivers and wetlands

**TABLE 1
THE CITY OF KIGALI URBAN SWAMPS PER DISTRICT**

No	Kigali City Districts	Land area (Ha)	Wetlands (Ha)	Wetland Buffer (Ha)	Percentage
1	Gasabo	429,146	2,826	736	8.28
2	Kicukiro	133,147	2,880	190	18.3
3	Nyarugenge	167,213	1,970	138	15
	Total/CoK	729,506	7676	1064	13.8

Source: Kigali Master Plan 2013

All activities have been grouped into 17 categories: residential home, commercial activities, industrial activities, parking, garages, ware houses, carpentry and welding workshops, dumping sites, bricks burning, petrol stations, carwash, schools, health centers, worship houses, domestic animal growing and play grounds.



FIGURE 3: Images of some illegal activities into the city of Kigali wetlands

TABLE 2
CATEGORIES OF ACTIVITIES DAMAGING WETLANDS IN THE CITY OF KIGALI

Activity	District			Total	Legal	Without legal Documents
	Gasabo	Kicukiro	Nyarugenge			
Residential home	1177	306	146	1629	703	926
Commercial activities	61	3	37	101	79	22
Industrial activities	6	73	5	84	78	6
Parkings	10	4	25	39	3	36
Garages	9	6	15	30	4	26
Ware houses	12	1	17	30	23	7
Carpentry and Welding Workshops	8	0	6	14	0	14
Dumping sites	4	2	10	16	0	16
Bricks burning	18	5	12	35	20	15
Petrol Stations	4	1	19	24	22	2
Carwash	2	0	4	6	0	6
Schools	2	2	2	6	2	4
Health centers	2	0	0	2	2	0
Worship houses	7	1	3	11	5	6
domestic animal growing	97	7	12	116	0	116
Play grounds	9	3	4	16	0	16
Garden centers	6	4	43	53	5	48

Source: Field survey

IV. DISCUSSIONS

Urban developments, including residential expansion and industrial and commercial activities, have contributed to the loss or degradation of Kigali's wetlands. The habitat's natural vegetation, dominated by *C. Papyrus* and a variety of *Pennisetum*, has been severely affected by industrial and household wastes. Industrial activities, such as brick making and sand mining, have contributed to the degradation of Kigali's wetlands.

For example, only parts of the Gikondo valley still fully function as a wetland because drainage and compaction from developments have degraded large parts of the area [8].

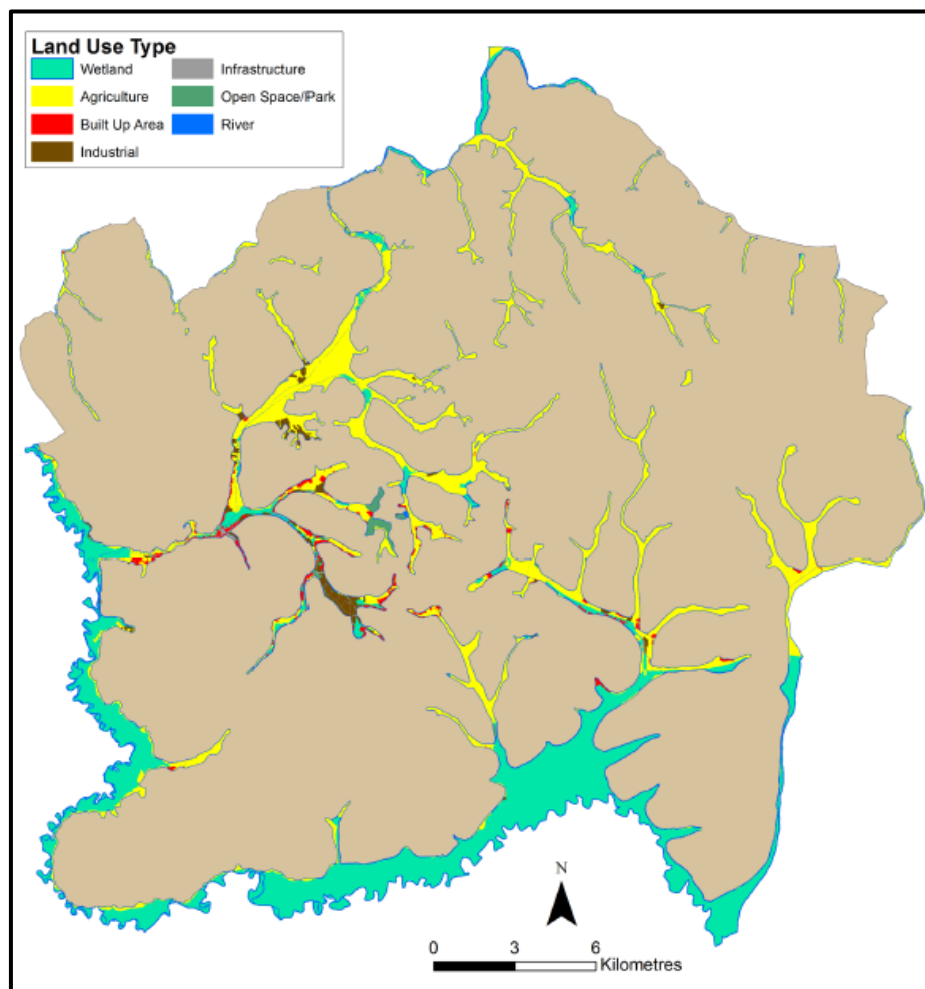


FIGURE 4: Types of land use currently in wetlands

Approximately 50 per cent of the wetlands in Kigali function as something other than a wetland (Fig. 3). Agriculture takes place in slightly over 39 per cent of wetlands. Built-up areas consisting of commercial buildings, public facilities and residences make up 2.7 per cent of wetlands. Green spaces, such as parks and rivers, account for less than 1 per cent. Over time, as sustainable city plans become implemented and constructed installations are removed from them, wetlands will start to regenerate [5].

Based on above findings, urban wetlands of the City of Kigali degraded drastically and their functions faced pressure from these activities. The consequences that are used to people of Kigali are several floods and the water pollution.

For years, a group of garages in Gatsata Sector, Gasabo District, known as the 'Gatsata Garages' were responsible for hazardous oils and heavy metals directly entering the Nyabugogo wetland because they lacked the proper infrastructure for waste disposal. Furthermore, most of the garage operators did not have toilets, making them dangerous to the natural environment and the health of members of the cooperatives operating in the neighborhood [9]. The garages' negative impacts on the wetland were so visually striking that the government moved quickly to relocate them to higher ground. Also, several garages that were affecting the wetland ecology, and whose businesses suffered during flooding events.

The article 19 of the Law N° 43/2013 of 16/06/2013 governing Land in Rwanda states that Swamp land belongs to the State. As stipulated by Article 116 of the Organic Law N° 04/2005 Of 08/04/2005 determining the Modalities of Protection, Conservation and Promotion of Environment in Rwanda states that the owners of the existing activities that do not respect the requirements in this organic law are obliged to respect the requirements of this organic law in a period not exceeding two (2) years from the day it comes into force [10].

The article 87 of the same law states that it is prohibited to construct houses in wetlands (rivers, lakes, big or small swamps), in urban or rural areas, to build markets there, a sewage plant, a cemetery and any other buildings that may damage such a place in various ways. All buildings shall be constructed in a distance of at least twenty (20) meters away from the bank of the swamp. But the law promulgated in 2005 was not respected.

As a result, people who have activities which were set up in an illegal way should relocate them to the suitable locations without compensation but those whose activities were set up in a legal way will be compensated for their relocation.

The relocation of all these activities from wetlands is imperative. Even if the operation would take long time but the political will is there, the law is clear and some activities started to be relocated like Gikondo Industrial Park that relocated to Kigali Special Economic Zone and Gatsata Garages that finished to be relocated.

After relocation of these activities, wetlands will be rehabilitated and surely, their recovery will be quick as some signs shown it at Gatsata.

Other wetland will be developed into recreational areas like Nyandungu that is being developed into recreational park.



FIGURE 5: some layout designs of Nyandungu recreational park (above is the Pope visit commemoration garden)

Some other projects for recreational purposes are being undertaken to preserve wetlands within the City of Kigali. Among them there is Kimicanga Entertainment Center in Gasabo District and Gikondo wetland parks.

V. CONCLUSION

The aim of this work was to present the will and commitment of the government of Rwanda for relocating all unnecessary activities from wetlands across the country and in Kigali particularly in the framework of environmental protection especially

for conservation of the existing swamps; to highlights different human activities established into wetlands of Kigali City and their categories; and to mention some initiatives of restoration that have started.

From our findings laws and regulations have been in place and this important as the government will. I addition, the action of identification of all activities into wetlands of the City of Kigali shows the high commitment in laws enforcement by the government. The potential polluters of wetlands have been identified and their implications in wetlands degradation.

All these activities will be relocated for the purpose of environmental protection in general, especially for conservation of the existing wetlands and protecting lives and businesses affected by the negative impact of their degradation. In this framework, people who have activities which were set up in an illegal way should relocate them to the suitable locations without compensation but those whose activities were set up in a legal way will be compensated for their relocation.

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The Biochemical Indices of Drought Resistant Species of Iori Plateau (East Georgia)

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Abstract— Existing forecasts of climate change predict significant warming, seasonal precipitation change, and strong and frequent droughts in the coming decades. Drought resistant plant species have more chance to survive. Predictions make the study of the biology of drought-resistant species especially relevant today. Antioxidant system, which plays an important role in plant stress resistance, is of special interest. Moreover, antioxidant substances are characterized by healing properties as well. Mechanisms of drought resistance of plants growing on arid territories of Georgia are practically unexplored. Presented study aimed to investigate the characteristics of antioxidant system of leaves of drought resistant species (*Euphorbia falcata* L. (sickle spurge), *Lycopsis orientalis* L. (small bugloss), *Cotinus coggygria* Scop. (smoke tree), *Elaeagnus angustifolia* L. (Russian olive) and *Amygdalus communis* L. (almond)) growing at one of the most arid regions of Georgia - Iori plateau (East Georgia). Analyses were made in two vegetative phases – flowering and fruit-bearing. From the obtained results, it is clear that the studied species have more or less different biochemical stress-adaptive mechanisms, which include certain enzymatic and non-enzymatic components of antioxidant system. In small bugloss in response to stress catalase was activated and synthesis of ascorbate-tocopherol and anthocyanins was enhanced; especially high amount of proline accumulation was noted. Phenols, anthocyanins and proline should be actively involved in stress resistance of sickle spurge. Russian olive was distinguished with high levels of ascorbate-tocopherol and anthocyanins, as well as proline; in addition the enzymatic antioxidants - catalase and peroxidase were activated, and soluble carbohydrates were accumulated. The protective systems of ascorbic acid and tocopherol, as well as phenolic compounds were active in smoke tree. From osmolytes content of proline increased, while the level of soluble carbohydrates was already the highest, compared to all tested species. Catalase was activated in response to stress in almonds; protective systems of ascorbate-tocopherol, phenols, and anthocyanins were active as well; among osmolytes content of carbohydrates was increased. The stability of carotenoids protective system of all studied species under stress conditions presumably indicates that experimental plants are less sensitive to radiation stress, and water deficiency is the main stress factor for them.

Keywords— antioxidants, drought resistance, osmolytes.

I. INTRODUCTION

Existing forecasts of climate change predict significant warming, seasonal precipitation change, and strong and frequent droughts in the coming decades (IPCC, 2007). It is likely that the combination of drought and unusually high temperatures that accompany global warming will cause vegetation change on Earth (IPCC, 2012; Jentsch *et al.*, 2007; Smith, 2011). The mentioned combination of drought and temperature forms the so-called "Global change type drought", also known as "hot drought". Under such drought conditions, the risk of severe plant stress and death is significantly increased (Allen *et al.*, 2015; McDowell *et al.*, 2008; Overpeck and Udall, 2010). Drought resistant plant species have more chance to survive.

Drought resistance is based on evolutionarily established physiological-biochemical mechanisms, many of which are studied today. Resistant plants are characterized by the high content of antioxidants and osmolytes. They possess the ability to alter metabolism in order to strengthen the antioxidant system, which helps to overcome the lack of water, as well as to recover from dehydration. (Aslam *et al.*, 2015; Laxa *et al.*, 2019).

Humans have been using plants for medicinal purposes since time immemorial. According to statistics more than 60% of the world's population today prefers medicinal plants and natural products. Moreover, 25% of the commercial preparations

produced in the world are of plant origin. In recent decades, interest in herbal remedies has grown in both Europe and America. This is due to the fact that phytopreparations are more available; furthermore, they are thought to have fewer side effects than synthetic drugs (Benzie and Wachtel-Galor, 2011; Harvey, 2000).

Against the background of expected climate change it may be assumed that the resources of plant raw material for medical use will be significantly reduced and probably replaced by drought-resistant species. All abovementioned predictions make the study of the biology of drought-resistant species especially relevant today. Characteristics of the antioxidant system, which play an important role in plant stress resistance, including drought, are of special interest. Moreover most antioxidant substances are characterized by healing properties as well (Pisoschi and Negulescu, 2011).

The physiological and biochemical indices of drought resistant plants growing on arid territories of Georgia are practically unexplored.

Based on the above, the aim of the study was to investigate the biochemical characteristics of leaves of the drought-resistant plants of one of the most arid regions of Georgia - Iori plateau (east Georgia). Mainly the elements of the antioxidant system have been studied: plastic pigments, ascorbic acid, tocopherol, anthocyanins, soluble phenols, total proteins, soluble carbohydrates, proline; as well as enzymes' - catalase, peroxidase and nitrate reductase activity, and total antioxidant activity, expressed in percents of inhibition.

Such studies give an idea of the adaptive mechanisms that either plant uses under stressful conditions and deepen our knowledge of the biology of drought-resistant plant species. This type of information under variable climate conditions allows to select plant species, that may be used against soil erosion and desertification, on the one hand, and as raw material for the preparation of natural remedies, on the other.

II. MATERIALS AND METHODS

2.1 Investigated species

Five species of plants have been studied: herbaceous - *Euphorbia falcata* L. (sickle spurge) and *Lycopsis orientalis* L. (small bugloss), woody plants - *Cotinus coggygria* Scop. (smoke tree), *Elaeagnus angustifolia* L. (Russian olive) and *Amygdalus communis* L. (almond). Healthy middle-aged leaves were collected for analysis from at least 5 different plants. The material was taken at an altitude of 828 m above sea level, at the territory of v. Udabno (Iori plateau, east Georgia), in two terms - in June (during the flowering phase) and in July (during the fruiting phase). Analyses were performed on raw material, with 3-fold repetition.

Sickle spurge (*Euphorbia falcata* L.) is one of the species of genus *Euphorbia*. It is an annual herbaceous plant from the family Euphorbiaceae; is distributed in semi-deserts, steppes and forest-steppes, is xeromesophyte. In general, sickle spurge grows in many parts of the world as a crop and ruderal weed (USDA, 2016). It is found on dry slopes and foothills in the Caucasus as well. The plant is used in folk medicine as a medicinal herb. Phytochemistry of members of the Euphorbiaceae family is currently being actively studied for the use of their compounds in medicine (Kemboi *et al.*, 2020).

Small bugloss (*Lycopsis orientalis* L.) is an annual herbaceous plant of the family Boraginaceae. The plant is considered as drought tolerant. It is found in the southern drought zone, where it grows in steppes, dry shrubs, roadsides, pastures. Small bugloss is a ruderal and agricultural weed; is used as food and in folk medicine. Currently its phytochemical composition is being studied more actively (Ertaş *et al.*, 2014).

Russian olive (*Elaeagnus angustifolia* L.) - a perennial shrub, or low-stemmed tree from Elaeagnaceae family. According to the literature, it can withstand both, very low (-30°C) and very high (+ 46°C) temperatures. Russian olive is a drought- and saline-resistant plant and plays an important role in protecting arid areas from erosion. It develops a strong root system that far exceeds the aboveground parts of the plant and is a strong water-absorbing mean. The lanceolate leaves of the plant are covered with scales and have a xeromorphic structure, which is an indicator of economic water consumption. During prolonged droughts, Russian olive loses 70% of its leaf cover and thus avoids water loss (Bartha and Csiszar, 2008). The plant is used in traditional medicine for healing (Tehrani-zadeh *et al.*, 2016).

Smoke tree (*Cotinus coggygria* Scop.) is a perennial shrub, from the family Anacardiaceae. The plant is less demanding to the soil - adapts to its different types and acidity in the range of 3.7-6.3. Smoke tree grows on dry, stony slopes, shrubs and forests; is light-requiring and drought-resistant; is used to plant and strengthen slopes (Global Invasive Species Database, 2020). The common area of smoke tree distribution includes the Caucasus, Central Asia, Central and Southern Europe, Asia Minor, Iran. The plant is widespread in Georgia, especially in its eastern part. It has long been used in folk medicine as a remedy (Matić *et al.*, 2016).

Almond (*Amygdalus communis* L.) is a tree from the Rosaceae family. It is accustomed to the climatic conditions of the Mediterranean, steppes and deserts, which are characterized by mild, humid winters and hot, dry summers. Almonds are light-requiring and drought-resistant, which is supported by a well-developed root system and economical transpiration (Kester *et al.*, 2003). The plant has long been used as a remedy for various diseases (Abdullah *et al.*, 2017).

2.2 Research area

The landscape of the Iori plateau varies from semi-desert to steppe and forest steppe. It is characterized by lack of water and scarce vegetation. The climate is dry, subtropical; the average temperature in January varies from 0 to -2°C, in July is 23°-24°C. The absolute minimum of temperature recorded in January was -13°C - -16°C, and the absolute maximum - in August was +37°C - +39°C. Precipitations of the plateau reach 499-600 mm per year, while in v. Udabno, where the experimental material was collected - 434 mm. Amount of precipitations increase from January and reaches its maximum in May, and then decreases (Aphkazava, 1975).

2.3 Biochemical assays

2.3.1 Antioxidant enzymes

Peroxidase activity was determined spectrophotometrically: optical density of the products of guaiacol oxidation was measured at the wave length of 470nm by the spectrophotometer (SPEKOL 11, KARL ZEISS, Germany) (Ermakov, 1987).

Catalase activity was studied gasometrically: volume of the oxygen released in the process of reaction between hydrogen peroxide and enzyme was measured (Pleshkov, 1985).

2.3.2 Nitrate reductase

Method of determining the nitrate reductase activity was based on measurement of nitrites amount, which were formed as a result of nitrate reductase reaction with the infiltrated nitrates (Ermakov, 1987).

2.3.3 Ascorbic acid

A titration method was used to measure the content of ascorbic acid in plant material. 2 g of fresh leaves were mashed in 15 ml of 2% hydrochloric acid and 10 ml of 2% metaphosphoric acid, and filtered. One ml of the filtrate was added to 25 ml of distilled water and titrated with a 0.001 M solution of dichlorophenolindophenole (Ermakov, 1987).

2.3.4 Tocopherol

Two g of ground leaves were extracted with 20-25ml of pure ethanol (three-fold). The combined extract was mixed with 20 ml of 60% potassium hydroxide, and saponificated on water bath for 2h. Tocopherol was extracted from the obtained hydrolyzate using diethyl-ether (3-fold extraction). The combined extract was washed with distilled water until a complete removal of alkaline residuals was detected by indicator paper. Water was removed with Na₂SO₄; the obtained solution was evaporated on the water bath, cooled, mixed with alcohol-nitric acid (1 ml of concentrated HNO₃:5ml of 96° alcohol), and boiled during 3 min till the color became dark red. Extinction of the extract was measured at 470nm by the spectrophotometer (SPEKOL 11, KARL ZEISS, Germany) (Filippovich *et al.*, 1982).

2.3.5 Anthocyanins

100mg of grinded leaves were added with 20 ml of 96% acidified (with 1% HCl) ethanol (99:1). After 24h retention in dark the optical density at 540nm was measured (spectrophotometer SPEKOL 11, KARL ZEISS, Germany) (Ermakov, 1987).

2.3.6 Plastid pigments

Chlorophylls and carotenoids were determined spectrophotometrically. Fresh leaves (100-200mg) were mashed with sand and CaCO₃ and washed with ethanol. Optical density of the filtrate was measured (spectrophotometer SPEKOL 11, KARL ZEISS, Germany). Concentration of chlorophylls a and b, also carotenoides was calculated by the formula of Wintermanns (Gavrilenko *et al.*, 1975).

2.3.7 Total phenols

A 0.5 g of fresh leaves was boiled in 80% ethanol for 15 min. After centrifugation the supernatant was saved, and residues of leaves were mashed in 60% ethanol and boiled for 10 min. Obtained extract was added to the first supernatant and evaporated. The sediment was dissolved in distilled water. One ml of the received solution was added with the Folin-Ciocalteu reagent and optical density was measured at 765 nm. The chlorogenic acid served as control (Ferraris *et al.*, 1987).

2.3.8 Total proteins

Content of proteins was determined after Lowry (1951).

2.3.9 Proline

0.5 g of dry leaves were mashed in 10ml of 3% sulphosalicylic acid and filtered. 2 ml of the filtrate was added to 2 ml of acid ninhydrin and 2 ml of ice acetic acid. After 1 h exposition on a water bath the extract was cooled and added with 4 ml of toluene and divided in a separating funnel. Optical density of upper layer was measured on a spectrophotometer (SPEKOL 11, KARL ZEISS, Germany) at 520 nm (Bates *et al.*, 1973).

2.3.10 Soluble carbohydrates

Content of soluble carbohydrates was tested with anthrone reagent (Turkina and Sokolova, 1971). To 100mg of air-dry leaf material was added 96° alcohol for extraction (3-fold). The total amount of the obtained extract was evaporated on a water bath and dissolved in 5ml of distilled water. To 0.5ml of the tested water extract was added 2ml of anthrone reagent and heated in a water bath for 10min. After this procedure the test-tubes were placed in a cold water bath and 15min later the optical density of the solution was measured at 620nm with a spectrophotometer (SPEKOL 11, KARL ZEISS, Germany).

2.3.11 Nitrates

After the water-extraction of 500g of plant material (homogenized for 30min at room temperature), it was filtered. Hydrogene peroxide was added to 10ml of the filtrate and evaporated. disulphophenolic acid was added to the obtained sediment and optical density was determined at 410nm (SPEKOL 11, KARL ZEISS, Germany) (Danilova, 1963; Pleshkov, 1985).

2.3.12 Total antioxidant activity

This index was measured by modified method using diphenyl-picryl-hydrazyl (DPPH) (Koleva *et al.*, 2002). 200 mg of experimental powder was extracted with 96° ethanol (two-fold). The obtained extract was evaporated on a water bath and the sediment was dissolved in 10ml of water-alcohol mixture. The 0.01ml of the received solution was added with 4ml of 40µM DPPH solution and after 30 minutes of incubation in the dark, the optical density was measured at 515nm by the spectrophotometer (SPEKOL 11, KARL ZEISS, Germany). The percent of inhibition was calculated.

2.3.13 Statistical processing of data

One way ANOVA and Tukey's multiple comparison tests were used to test differences between the means. All calculations were performed using statistical software Sigma Plot 12.5.

III. RESULTS AND DISCUSSION

As mentioned above, the study of biochemical characteristics in the leaves of experimental plants was carried out in June (flowering phase) and July (fruiting phase). In June, the soil on the Iori plateau is not very dry, as it still retains traces of May precipitations; Plants therefore experience less drought pressure, despite strong insolation and high temperature. While in July, when the plants are in fruiting phase, the soil becomes very dry due to lack of precipitations; the high temperature and intensive irradiation combined with water deficiency establish a very stressful environment for existence. Nevertheless, the experimental species continue to vegetate, that is indication to their resistance to existing stress conditions.

Investigation of the studied characteristics in June and July allowed following their change under water deficit conditions and in two different phases of vegetation. The results obtained according to the vegetation phases within one species are compared with each other, as well as the data of different species in each phase.

3.1 Catalase and peroxidase

Catalase and peroxidase, along with other antioxidant enzymes, catalyze the detoxification of superoxide radicals and hydrogen peroxide in plants (Garifzyanov *et al.*, 2011). It has been established that in drought tolerant species mostly peroxidase, ascorbate peroxidase and catalase are activated to neutralize active forms of oxygen (Kapoor *et al.*, 2020; Laxa *et al.*, 2019).

As catalase is mainly concentrated in peroxisomes, it actively contributes to the neutralization of hydrogen peroxide generated during photosynthesis, which is synthesized in large quantities under water-deficient conditions (Noctor *et al.*, 2014). It has been established that more than 70% of hydrogen peroxide generated under drought conditions is the result of photorespiration (Cruz, and de Carvalho, 2008).

From the obtained results it is clear that the activity of catalase in fruiting phase increased 4.9 times in small bugloss leaves and 1.2 times in Russian olive and almond leaves, compared to flowering phase ($p \leq 0.001$, $p = 0.01$ and $p = 0.03$, respectively); while in smoke tree leaves it decreased by 1.6 times ($p = 0.02$), and remained statistically same in sickle spurge ($p = 0.7$) (Fig. 1).

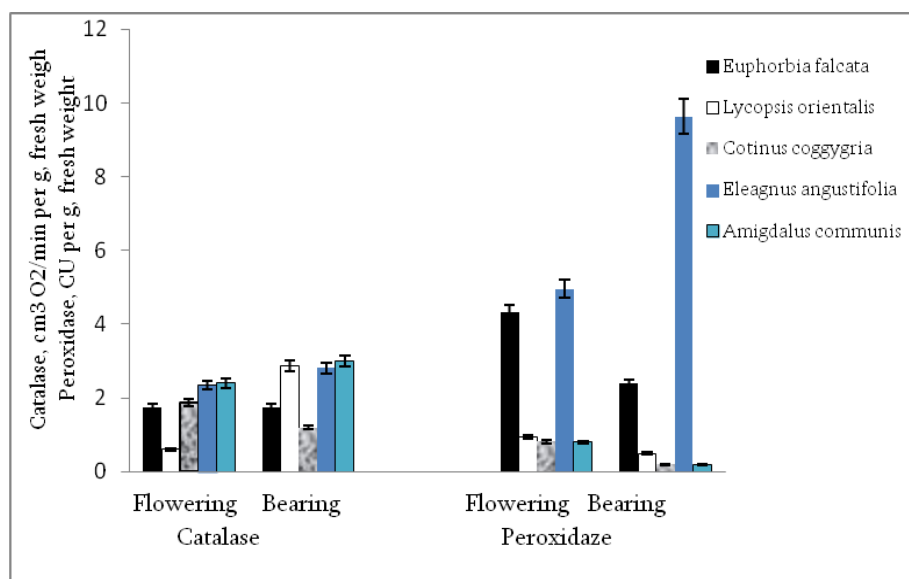


FIGURE 1: Activity of catalase and peroxidase in leaves of drought resistant species of Iori plateau (East Georgia) plants in two vegetative (flowering and fruit-bearing) phases

Comparison the results between species has revealed that the catalase activity in flowering phase in sickle spurge and smoke tree ($p = 0.8$) as well as in Russian olive and almond ($p = 0.9$) was statistically similar. In fruiting phase, statistically similar results were found in small bugloss, Russian olive and almond leaves ($p \geq 0.06$) (Fig. 1).

Catalase is believed to work more actively under conditions of high concentrations of hydrogen peroxide (Nyathi, and Baker, 2006). Taking this fact into account, it can be assumed that one of the consequences of stressors aggravation in July was the high concentrations of hydrogen peroxide in small bugloss, Russian olive and almond leaves, which induced the activation of catalase antioxidant mechanism in these plants.

Peroxidases are multifunctional enzymes that use various reducing agents, most commonly phenolic compounds, to neutralize hydrogen peroxide. The involvement of peroxidases in antioxidant protection of various plants under the drought has been shown (Kolupaev, Kokorev, 2019).

Peroxidase activity statistically reduced in leaves of all studied species during the fruiting phase, (1.8 times in sickle spurge, 1.9 times in small bugloss, 4 times in smoke tree and almonds ($p \leq 0.003$)), except Russian olive (the index in leaves of the latter increased 1.9-times, $p \leq 0.001$) (Fig. 1).

Significant increase in peroxidase activity in Russian olive leaves, where by less intensity but increase in catalase activity also has been observed, suggests that the enzymatic antioxidant system in this species is one of the most effective means of protection against drought and other stressors.

Peroxidase activity in the flowering phase was found to be statistically similar in small bugloss, smoke tree and almond leaves ($p \geq 0.3$); while in the fruiting phase statistically similar and the lowest data were observed in smoke tree and almond leaves ($p > 0.05$).

Thus, based on the results obtained, it may be concluded that the enhanced water deficiency in July, accompanied by the intensive insolation and high temperature, activates the catalase-peroxidase defense enzymatic mechanism in Russian olive, while in small bugloss and almond the catalase protective mechanism is more active.

It is believed that despite the important role of enzymes in the detoxification of active forms of oxygen, the enzymatic antioxidant system cannot provide complete protection of the cell from damage, which is due to a number of factors (Polesskaya *et al.*, 2006). Therefore, it has been suggested that low-molecular-weight antioxidants, such as ascorbic acid,

tocopherol, anthocyanins, phenolic compounds, etc., often more effectively protect metabolism from active forms of oxygen (Blokhina *et al.*, 2003; Radyukina *et al.* 2012).

3.2 Ascorbic acid and tocopherol

Ascorbic acid is one of the most important low molecular weight antioxidants synthesized in the plant. It affects many physiological processes. However, its main role is to protect plant metabolic processes from free radicals. Ascorbic acid is involved in the recovery of tocopherols and in the xanthophyll cycle; Thus, it also participates in the protection of the photosynthetic apparatus and membranes in general (Shao *et al.*, 2007; Smirnov, 2000). Ascorbic acid is found in various organelles and apoplasts of plant cells. It is most abundant in chloroplasts (20–300 mmol) (Horemans *et al.*, 2000; Smirnov, 2000).

During the fruiting phase the ascorbic acid content in small bugloss and smoke tree leaves increased 2.2 and 1.6 times, respectively ($p \leq 0.001$) (compared to flowering); in sickle spurge leaves - decreased by 8.5% ($p = 0.012$), while in Russian olive and almond leaves the data of both phases were statistically similar ($p = 0.6$ and $p = 0.25$, respectively) (Fig. 2).

Comparing the data of experimental species have revealed that in the flowering phase the content of ascorbic acid in sickle spurge and smoke tree leaves was statistically similar ($p = 0.09$), while in other species it was statistically different ($p < 0.05$). The maximal content of vitamin was determined in leaves of Russian olive and the minimal - in leaves of small bugloss (Fig. 2).

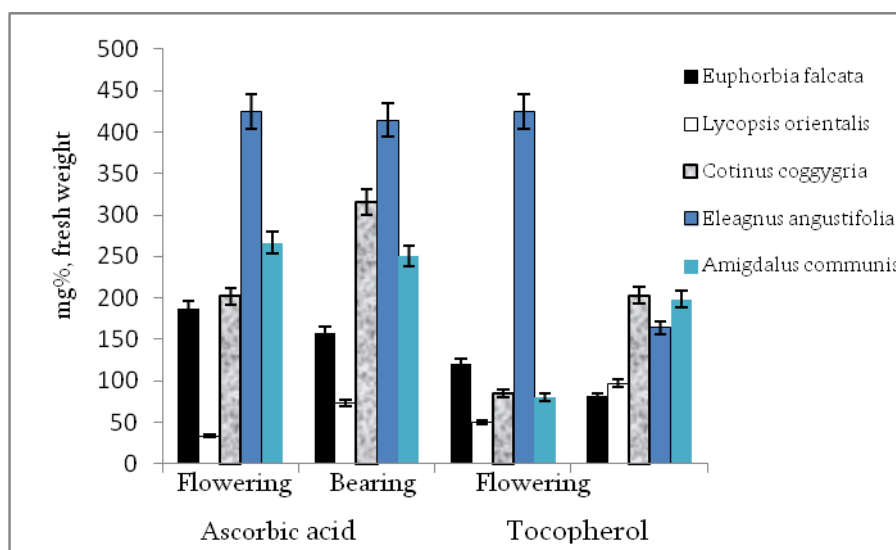


FIGURE 2: Content of ascorbic acid and tocopherol in leaves of drought resistant species of Iori plateau (East Georgia) plants in two vegetative (flowering and fruit-bearing) phases

The ascorbic acid content in leaves of all experimental plants was statistically different during the fruiting phase ($p < 0.001$).

Increase and maintenance of the ascorbate content in the cell is considered as one of the indicators of plant resistance to stress (Kolupaev and Kokorev, 2019). Enhancement of the ascorbic acid under the conditions of intensive radiation and drought has been established (Yang *et al.*, 2008). It affects the activity of many enzymes and reduces oxidative stress-induced damage through synergistic interactions with other antioxidants. It is believed that ascorbate-dependent protective system is activated in drought-resistant plants (Pourcel *et al.*, 2007).

According to significant increase of ascorbate in small bugloss and smoke tree leaves under the stress conditions, it may be suggested that it is one of the leading antioxidants in the drought resistance of these plants.

Tocopherol content increased 1.9 times in small bugloss leaves, 2.4 times in smoke tree and 2.5 times in almond leaves (compared to flowering) ($p = 0.008$, and $p \leq 0.001$) during the fruiting phase; while decreased in sickle spurge and Russian olive leaves 1.5 and 2.6 times, respectively ($p = 0.031$ and $p \leq 0.001$) (Fig. 2).

In June (flowering phase) the maximal amount of tocopherol was found in Russian olive compared to other studied species. The vitamin content measured in the leaves of smoke tree was statistically similar to the data established in leaves of sickle

spurge, small bugloss and almond ($p > 0.05$). Different results were obtained in sickle spurge and small bugloss leaves ($p = 0.003$) (Fig. 2).

In July (fruiting phase) statistically similar results of tocopherol content were obtained in leaves of sickle spurge and small bugloss ($p = 0.5$), smoke tree and Russian olive ($p = 0.06$), smoke tree and almond ($p = 0.9$), Russian olive and almond ($p = 0.05$) (Fig. 2)

Tocopherols are antioxidant compounds found in all parts of the plant (Srivalli *et al.*, 2003). They are the most active and dominant antioxidants in the chloroplast membrane and mainly serve as protectants against photooxidation. (Ledford and Niyogi, 2005; Mullineaux *et al.*, 2006). Especially active is α -tocopherol, the content of which increases in the photosynthetic tissue in response to various abiotic stresses (Abbasi *et al.*, 2008; Giacomelli *et al.*, 2007; Noctor, 2006). Increase of tocopherols in combination with ascorbic acid is one of the plant's primary responses to water deficiency (Hasanuzzaman *et al.*, 2014; Pourcel *et al.*, 2007; Wu, *et al.*, 2007). Therefore, it can be concluded that tocopherols, along with ascorbic acid, are an important factors in drought resistance of small bugloss, smoke tree and almonds. Since the content of ascorbic acid in leaves of Russian olive did not change during the fruiting phase, and the amount of tocopherol even decreased, the pool of these compounds in leaves was still higher compared to all the studied species, which is the indication to the important role of these compounds in the drought resistance of Russian olive as well.

3.3 Soluble phenols

Phenolic compounds are a large group of secondary metabolites that play an essential role in vital processes of a plant. These compounds have a strong ability to bind free radicals and effectively protect the cell membrane from damage caused by oxidative stress (Winkel-Shirley, 2002). Under conditions of abiotic stress, including water deficiency, the biosynthesis of phenolic compounds in the plant increases. A number of literary data demonstrate the accumulation of phenolic compounds in plants during drought; which significantly increased their drought resistance (Sharma *et al.*, 2019).

The content of soluble phenols in leaves of the studied species was not high, except smoke tree, which is known for its particularly high content of these compounds (Matić *et al.*, 2016).

From the results obtained, it is clear that the response to the enhancement of stressors in experimental plants in July was different in terms of the content of phenolic compounds; including those species that demonstrated statistically similar content of phenolic compounds in the flowering phase (sickle spurge, Russian olive, almond) (Fig. 3). During the fruiting phase, the content of phenolics in sickle spurge and almonds increased 3-fold and 1.6-fold, respectively (compared to the flowering phase ($p \leq 0.001$)), while in smoke tree and Russian olive decreased by 14% and 20%, respectively ($p < 0.05$); In small bugloss, which was distinguished by the minimal content of phenols compared to other species, the index did not change statistically ($p > 0.05$) (Fig. 3).

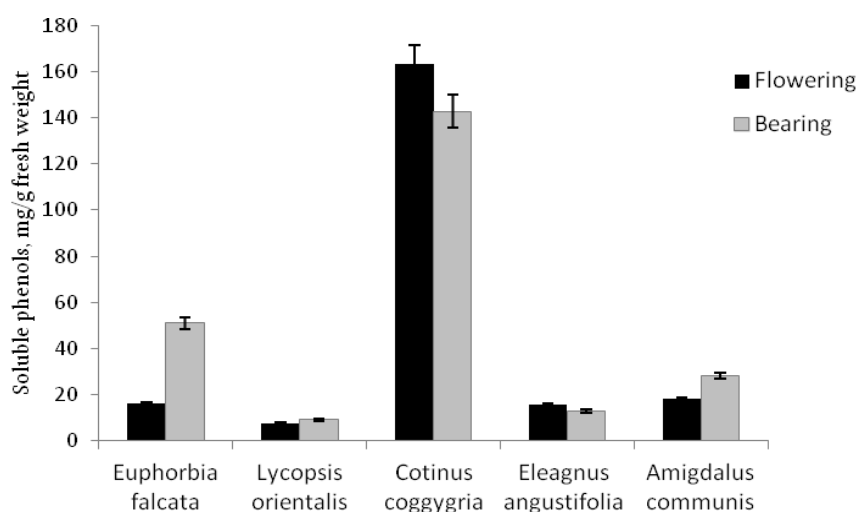


FIGURE 3: Content of soluble phenolic substances in leaves of drought resistant species of Iori plateau (East Georgia) plants in two vegetative (flowering and fruit-bearing) phases

While comparing species, in June statistically similar results were revealed in sickle spurge, Russian olive and almonds; results of all species in July were statistically different (Fig. 3).

Thus, the antioxidant system of sickle spurge and almonds responded to the enhanced water shortage and elevated temperature in June by the activation of phenylpropanoid pathway. Taking into account the abatement of ascorbate-tocopherol pool in sickle spurge under the enhanced stresses, the pronounced activation of phenolic synthesis may be considered as one of the leading mechanisms of stress adaptation in this species. Despite a slight decrease in phenolics content in smoke tree in July, its especially high pool of these compounds evidently plays an essential role in resistance to drought and temperature stresses. Here must be added the fortified ascorbate-tocopherol system, which “protects” smoke tree, as well as it happens in small bugloss.

3.4 Plastic pigments and anthocyanins

The functional state of the photosynthetic apparatus is assessed by the content of chlorophylls and carotenoids (Lichtenthaler and Buschmann, 2001). At the same time it is a good indicator to evaluate the performance of the antioxidant system.

The content of chlorophylls in leaves of the experimental plants changed with different regularities in June-July. The results demonstrate that chlorophylls content in the fruiting phase increased by 25% in sickle spurge ($p=0.01$), in almonds - did not change statistically ($p=0.8$), while in other species - decreased ($p<0.05$): in small bugloss – by 25.5%, in smoke tree - by 28%, and in Russian olive - by 11% (compared to the flowering phase). Smoke tree and Russian olive leaves were distinguished by high chlorophyll content among the studied species (Fig. 4).

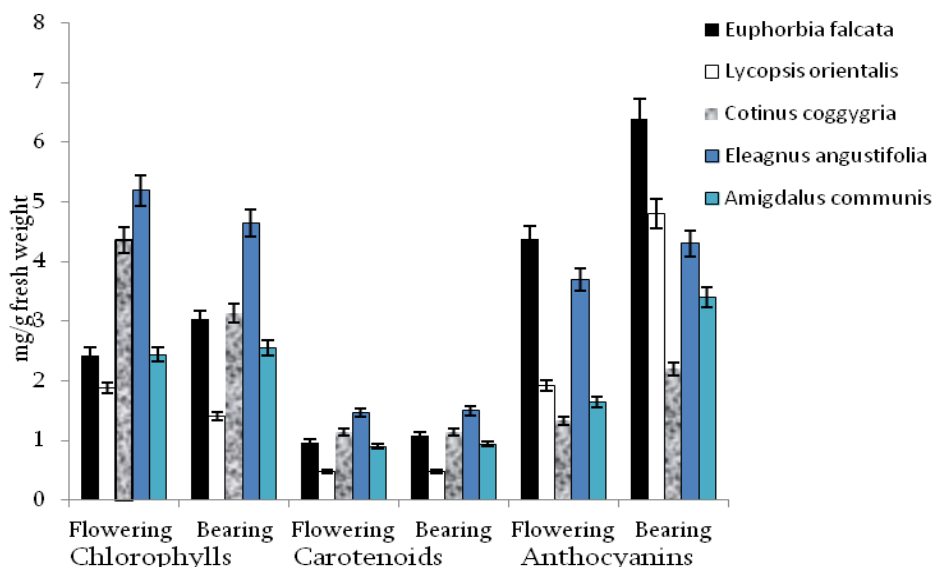


FIGURE 4: Content of chlorophylls, carotenoids and anthocyanins in leaves of drought resistant species of Iori plateau (East Georgia) plants in two vegetative (flowering and fruit-bearing) phases

Comparison of species by chlorophylls content, revealed statistically similar results in sickle spurge and almonds in the flowering phase ($p=0.4$), and in sickle spurge and smoke tree in the fruiting phase ($p=0.9$). At the same time, by the maximal content of both chlorophylls and carotenoids were distinguished leaves of Russian olive, and by the minimal - those of small bugloss (Fig. 4).

Chlorophyll reduction is a common event under drought conditions; which may be caused by inhibition of pigment biosynthesis as a result of stress, or breakdown of pigments (Batra *et al.*, 2014). It has an adaptive purpose and relieves excess energy of the plant photosynthetic apparatus. In particular, under conditions of water shortage and high temperature, when the plant is forced to close its stomata to prevent desiccation, the efficiency of photosynthesis decreases and besides the energy absorbed by the plant remains unused; it also causes the accumulation of active forms of oxygen. The last of its side poses a risk of oxidative stress in the plant (Noctor *et al.*, 2014). Avoidance of this phenomenon is achieved by the destruction of pigments, i.e. by reducing their number (Mafakeri *et al.*, 2010). Chlorophyll is constantly synthesized and decomposed under light conditions; but under intensive illumination, accompanied by high temperatures, the rate of its decomposition exceeds the synthesis and the equilibrium shifts towards low pigment concentrations (Morais *et al.*, 2007).

Thus, the reduction of chlorophylls in small bugloss, Russian olive and smoke tree should be considered as an expression of adaptation to stressful conditions along with the intensification of drought. It may be assumed that the above mentioned equilibrium between the synthesis and decomposition of chlorophyll under stress conditions should not fall below a certain

minimal level of chlorophyll content for the plant to continue and complete its vegetation cycle. At the same time, no "excess" amount of chlorophyll is needed, because the plant not only "is not able to use" it, but it is also "harmful" to it. Indeed, if we observe the chlorophyll content in the studied plants, by its highest content in the flowering phase were distinguished Russian olive and smoke tree. Such "excess" pigments posed a "threat" to these plants in the face of drought. Consequently, they decreased during the fruiting phase, although less in the leaves of Russian olive. The last can be related to the anatomical features of the leaves of this plant: they are covered with scales, which protect them from excess light and overheating (Fig. 4).

After decrease in July, the chlorophyll content in smoke tree leaves became statistically equal to the increased content of the pigment in sickle spurge leaves. It may be assumed that this amount of chlorophyll was within the "safe limits" for plants under those light, temperature, and water supply conditions, which were on the studied area in July; so it became "permissible" for sickle spurge to increase its chlorophyll content despite stress enhancement. Here should also be noted one advantage of the sickle spurge photosynthetic apparatus in its resistance to stress: it is established that the species of *Euforbia* genus have C_3 photosynthesis under the conditions of better water supply and relatively low temperature, while under high temperature and water deficiency they "switch to" C_4 or CAM metabolism, which allows to fix more carbon through reduced water loss and thus they are better adapted to hot and dry conditions (Batanouny *et al.*, 1991; Davies *et al.*, 2018).

It may be assumed that such an event occurs in the species of studied sickle spurge, which corroborates its greater adaptation to the conditions of increased stress.

As for almonds, the chlorophylls data in its leaves were statistically similar in June and July and quantitatively approached the June rate of sickle spurge. It seems that for almonds this number of chlorophylls, as for the tree plant, was "sufficient" to survive under stress (Fig. 4).

In small bugloss, which revealed the lowest chlorophyll content in leaves, the pigment amount fell by another 25% in July; however, this did not prevent him, as an annual herb, to complete his life cycle successfully. It turns out that the reduced level of chlorophylls was "enough" for him.

While discussing about the content of chlorophylls, of course, it should not be forgotten their protective and auxiliary pigments - carotenoids. These compounds with antioxidant properties belong to different groups of terpenoids, which play an important role in protecting the photosynthetic apparatus from excess light and are responsible for its structural integrity in the thylakoid membrane (Guidi *et al.*, 2017). However, there is another group of carotenoids called secondary carotenoids, which are not structurally related to the photosynthetic apparatus and accumulate outside the thylakoids. They play an active role in protecting cell structures from photo-damage as potent antioxidants, by neutralizing active forms of oxygen and enhancing the sink ability of photoassimilates. Thus, the photoprotective mechanism based on secondary carotenoids does not require additional energy and metabolites and is favorable for the plant to adapt to long-term stress, will it be an excessive radiation, drought, extreme temperatures, or salinization (Solovchenko and Neverov, 2017).

Carotenoids content in experimental species did not change statistically by phases ($p > 0.05$) (Fig. 4). Comparison of species with each other revealed that both in June and July statistically similar carotenoids content was in sickle spurge, smoke tree and almond leaves ($p > 0.05$). It should be noted that the difference in carotenoid content between the studied species was similar to the chlorophyll content, i.e. the higher was the carotenoid content in leaf, the higher was its chlorophyll content, which should be related to the protective function of carotenoids; This may explain the low chlorophyll content of small bugloss, the similar chlorophyll values of almond, sickle spurge, and smoke tree leaves, or even the maximal chlorophyll content of Russian olive leaves (Fig. 4).

Another group of pigments with antioxidant properties that accumulate in the vegetative tissues of plant in response to various abiotic stresses are anthocyanins - compounds of the flavonoid group (Gould, 2004). In July, during the fruiting phase, when the plants were exposed to stronger drought, solar irradiation, and high temperatures, the anthocyanins content in leaves of most experimental plants increased significantly, compared to June data. Sickle spurge, which had the highest content of anthocyanins in June, showed a 46% increase in pigments ($p = 0.006$); the content of anthocyanins increased 2.5 times ($p = 0.009$) in small bugloss leaves and 2 times - in almond leaves ($p = 0.001$). No statistical difference was found between the two-phase data of smoke tree and Russian olive leaves ($p = 0.07$ and 0.15 , respectively) (Fig. 4).

Between-species comparison of anthocyanins content it was found that in the flowering phase this data of sickle spurge and smoke tree did not differ statistically ($p = 0.3$) and were maximal. Statistical differences between other species were also not established ($p > 0.05$) (Fig. 4).

In the fruiting phase, sickle spurge was again mentioned with the highest content of anthocyanins compared to other experimental species. The minimal data were revealed in smoke tree, while in small bugloss, Russian olive and Almond the data were statistically similar ($p>0.05$) (Fig. 4).

The synthesis of anthocyanins in vegetative tissues enhances in response to various abiotic stresses, including drought, excessive light, and unfavorable temperatures. They bind active forms of oxygen and reveal photoprotective, and signaling function under stress (Gould *et al.*, 2018; Kovinich *et al.*, 2015). Significant increase in anthocyanins in sickle spurge, small bugloss and almond leaves against the background of increased stress makes to suggest that these pigments make an important contribution to the protection of the studied plants from oxidative stress, including their photosynthetic apparatus.

3.5 Amino acid Proline, total proteins and soluble carbohydrates

In addition to hydrogen peroxide converting enzyme systems, many metabolites may be involved in its neutralization, which reveal antioxidant properties but are not antioxidants. In particular, synthesis and accumulation of osmoprotectants, such as free amino acids, proteins, and soluble sugars, has been established under stress in plants. They protect membrane protein-lipid components from denaturation. (Franco and Melo, 2000; Iqbal *et al.*, 2020; Meng, 2014; Szabados and Savory, 2010).

The role of proline as an osmoregulator in plant adaptation to drought is well known. Numerous papers have shown the positive impact of proline accumulation on drought resistance (Ashraf *et al.*, 2018; Kaur and Asthir, 2015). It increases the cell osmolarity which in turn attracts water to the cell, or reduces its outflow; thus providing the water potential to maintain turgor in the cell under conditions of water deficiency (Anjum *et al.*, 2000; Joseph *et al.*, 2015; Kartashov, 2013). The last decade has focused on the antioxidant effect of proline. A model is proposed that demonstrates the ability of proline to neutralize hydroxyl radicals (Signorelli *et al.*, 2014).

Among the studied species small bugloss and Russian olive were distinguished with high content of proline in the flowering phase. During the fruiting phase, the index increased by 15% in sickle spurge and smoke tree leaves ($p=0.004$ and $p=0.009$, respectively), while it decreased in leaves of small bugloss, Russian olive and almond, by 11.4%, 11.7% and 38.4% respectively ($p\leq 0.001$, $p=0.01$, and $p=0.004$) (Fig. 5).

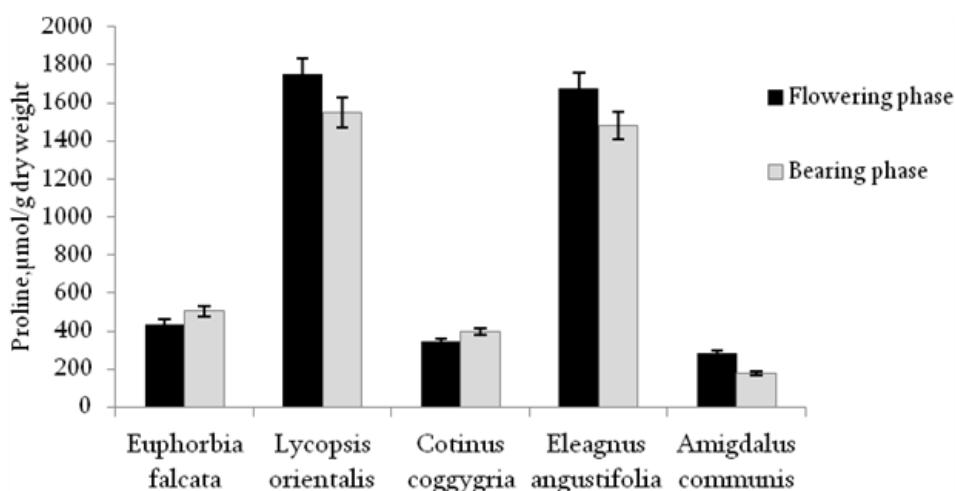


FIGURE 5: Content of proline in leaves of drought resistant species of Iori plateau (East Georgia) plants in two vegetative (flowering and fruit-bearing) phases

According to June results, the proline content in the experimental species was statistically different ($p<0.05$), while in July only in small bugloss and Russian olive leaves were statistically similar ($p=0.07$) (Fig. 5).

At first glance, it may seem that in most of the species studied, the proline accumulation strategy is not the leading one against water deficiency. A slight increase in this compound in sickle spurge and smoke tree leaves makes to suggest that it plays some role in protecting these species from stress; but if we observe how high the proline content is in leaves of small bugloss and Russian olive, despite its slight decrease in July, we must assume that this compound plays one of the leading roles in the drought resistance of these species.

Thus, in sickle spurge, smoke tree and especially small bugloss and Russian olive leaves proline should be considered as one of the means of protection from drought.

It is known from the literature that adaptation to unfavorable environmental conditions is accompanied by changes in the qualitative and quantitative composition of proteins (Mohammadkhani and Heidari, 2008; Parida *et al.*, 2007).

The quantitative study of total proteins showed that in the fruiting phase their content decreased significantly in all experimental species ($p \leq 0.001$) (compared to the flowering phase): in sickle spurge and small bugloss - 2 times, in smoke tree - 1.6 times, in Russian olive - 1.4, 1.5 times (Fig. 6).

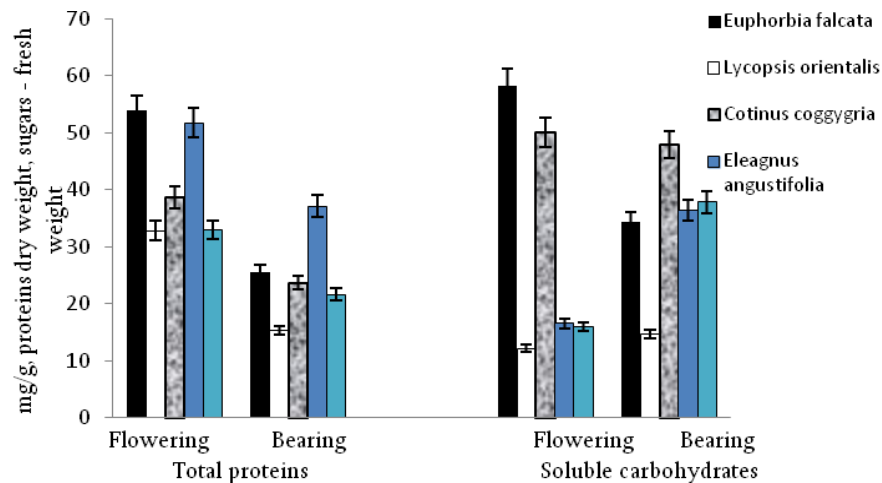


FIGURE 6: Content of total proteins and soluble carbohydrates in leaves of drought resistant species of Iori plateau (East Georgia) plants in two vegetative (flowering and fruit-bearing) phases

When comparing the results between species, the highest content of total proteins was found in sickle spurge and Russian olive leaves. In June the index was statistically similar in small bugloss - almonds ($p=0.5$), and sickle spurge - Russian olive ($p=0.4$), in July - in sickle spurge and smoke tree ($p=0.05$) (Fig. 6).

Decrease in total proteins under various stresses has been shown, which may be related to the outflow of soluble nitrogen-containing compounds from the leaves or their reduced synthesis (Sorkheh *et al.*, 2012). At the same time, the reduction of total proteins under drought conditions is explained by reduced photosynthesis, on the one hand and proteolysis of proteins, on the other (Mohammadkhani and Heidari, 2008; Taiz *et al.*, 2016). In the case of tested species none of the above mentioned reasons is excluded. In most experimental plants a decrease in chlorophylls occurred during the fruiting phase, which implies a decrease in photosynthetic activity, while an increase in the content of the amino acid proline, as an osmoprotectant, in some species may be the result of proteolysis.

The decrease in total protein content in leaves of experimental plants may be partially related to the developmental phase as well. In the fruiting phase, when the fruit is growing and forming, the flow of assimilates towards it increases, as to an important acceptor. This is reflected on the leaf composition as a donor as well (Tegeger and Masclaux - Daubresse, 2018).

It has been established that synthesis of so called stress proteins are activated in plant cells under various stress conditions, including drought. These proteins, known as dehydrins and exerting an osmolite-like effect, are involved in the stabilization and osmotic regulation of membrane proteins. At the same time they regulate the concentration of solutions in the cytoplasm and protect cell structures from oxidative stress (Iqbal *et al.*, 2020; Mohammadkhani and Heidari, 2008). Although dehydrins have not been the subject of our study, we must assume that they would inevitably be in the leaves of experimental plants as in drought-resistant species. Their study is the subject of further research.

Although soluble carbohydrates are involved in the metabolic pathways that lead to the formation of active forms of oxygen, they also play an important role in neutralizing oxygen. The high sugar content protects proteins from denaturation and promotes membrane stabilization (Couee *et al.*, 2006; Laxa *et al.*, 2019).

A number of papers demonstrates the accumulation of soluble carbohydrates in plants in response to various stresses (Finkelstein and Gibson, 2001; Mohammadkhani and Heidari, 2008; Prado *et al.*, 2000). Carbohydrates create a low water potential in the cell, which is necessary for the absorption of water from a highly mineralized soil solution (Eryomchenko *et al.*, 2013).

High carbohydrate content in the flowering phase was found in sickle spurge and smoke tree leaves, compared to other experimental species. In the fruiting phase the index decreased 1.7 times in sickle spurge ($p=0.001$), in Russian olive and almond increased 2.2 times and 2.4 times ($p=0.002$ and $p<0.05$) respectively, while in small bugloss and smoke tree it was statistically similar to the previous phase ($p=0.09$ and $p=0.7$) (Fig. 6).

Comparing the results between species clears that all data in June statistically differed from each other ($p < 0.001$). Statistically similar results were obtained in July in Russian olive and almonds ($p=0.05$) (Fig. 6).

Significant increase in soluble sugars under stress conditions in Russian olive and almond leaves makes to suggest that these osmolytes should play a leading role in protecting these plants from drought stress. Generally high content of soluble sugars in smoke tree leaves also demonstrates a stress-protective function of these compounds.

3.6 Nitrate reductase and nitrates

Nitrogen metabolism, growth and productivity of the plant can be discussed according to nitrate reductase (NR, E.C. 1.6.6.1) activity, as it plays an important role in the regulation of these processes (Garg and Singla, 2005). During the flowering phase, a high rate of nitrate reductase activity was observed in small bugloss, while in smoke tree and almond the enzyme's activity was not detected at all (Fig. 7).

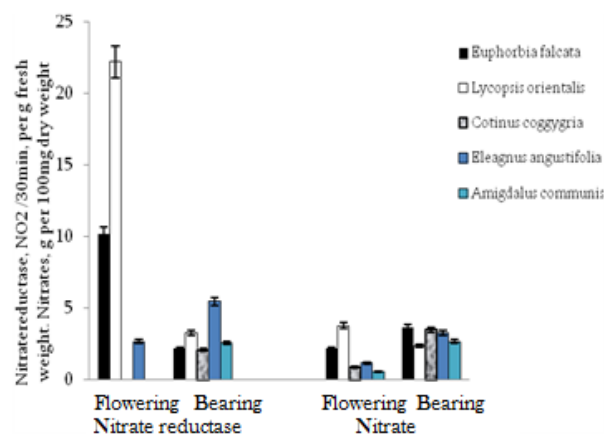


FIGURE 7: Nitrate reductase activity and nitrates content in leaves of drought resistant species of Iori plateau (East Georgia) plants in two vegetative (flowering and fruit-bearing) phases

In the fruit bearing phase, a sharp decrease in nitrate reductase activity was observed in sickle spurge and small bugloss (4.6 and 6.7 times, respectively; $p \leq 0.001$), while in smoke tree and almonds, on the contrary - nitrate reductase activity exceeded zero. Enzyme' 2-fold activation was observed in Russian olive leaves ($p=0.002$) as well (Fig. 7).

Comparison of species revealed that during the flowering phase enzyme; activity statistically differed in all tested variants ($p < 0.001$); while in the fruit bearing phase only Russian olive was distinguished by statistically different ($p < 0.001$) results (Fig. 7).

Decrease in the activity of nitrate reductase under drought conditions has been established, which is associated with a decline in photosynthetic activity (Kapoor *et al.*, 2020).

Experimental results on nitrate reductase activity demonstrate fairly a strange picture, which may be related to the life-forms of the experimental species: sickle spurge and small bugloss are therophytes, while smoke tree, Russian live and almond are phanerophytes. The conversion of nitrates in herbaceous plants with the participation of nitrate reductase actively takes place in leaves. While it is known that in a number of woody plants the reduction of nitrates occurs mainly in roots (Cruz *et al.*, 1993; Thomas and Hilker, 2000). Accordingly, the enzyme was active in sickle spurge and small bugloss leaves during the flowering phase, although during the fruiting phase, its activity sharply reduced under stress conditions (Fig. 7). The fact that nitrate reductase is a substrate-dependent enzyme should be taken into account as well. This means that its synthesis and activity are regulated by the concentration of nitrates and intracellular ammonium (Dias *et al.*, 2011; Nicodemus *et al.*, 2008). Nitrates content in smoke tree and almond leaves was particularly low during the flowering phase, which probably led to a delay in nitrate ductase activity (Fig. 7).

Nitrate reductase activity is also inhibited by light, if carbon fixation is impaired (Kaiser and Forster, 1989). Such conditions are created by water shortage. In July, plants were exposed to such stressful conditions, which were reflected on enzyme' decreased activity (Fig. 7).

Regulation of nitrogen metabolism plays an important role in plant stress resistance, as many physiological processes are associated with it. Drought inhibits nitrogen uptake and movement to aboveground parts of the plant, leading to reduced transpiration and cell membrane permeability. Due to the soil drying it is also difficult to dissolve nitrogen and ammonium salts in water. It turned out that the activity of antioxidant enzymes significantly depends on the nitrogen concentration. Therefore, under conditions of poor nitrogen supply, the effect of oxidative stress on the plant enhances (Iqbal *et al.*, 2020).

The highest content of nitrates was detected in small bugloss leaves in the flowering phase. However, during the fruit-bearing phase, this index decreased 1.6 times ($p=0.014$). In small bugloss in the flowering phase in terms of the high content of nitrates the activity of nitrate reductase was highest; which decreased in the fruiting phase in parallel with the decrease in nitrates content. In the fruiting phase, the content of nitrates in leaves of analytical plants significantly increased: in sickle spurge - 1.9 times, smoke tree - 3.9 times, Russian olive - 2.7 times, in almonds - 2.5 times ($p<0.05$). This accordingly, caused an increase in nitrate reductase activity in woody species (Fig. 7).

Comparison of nitrates content between species showed that in the fruit-bearing phase the index was statistically similar in all studied species ($p>0.1$) (Fig. 7).

3.7 Total antioxidant activity

This is an integrated characteristic of the ability of hydrophilic antioxidants to bind free radicals, without specifying compounds (Arnao *et al.*, 1999). Investigation of the total antioxidant activity of tested leaves has shown that during the fruiting phase the index increased in sickle spurge and small bugloss 1.9 and 1.3 times respectively. In smoke tree and Russian olive it stayed unchanged, while in almond - decreased by 1.3 times (Fig. 8).

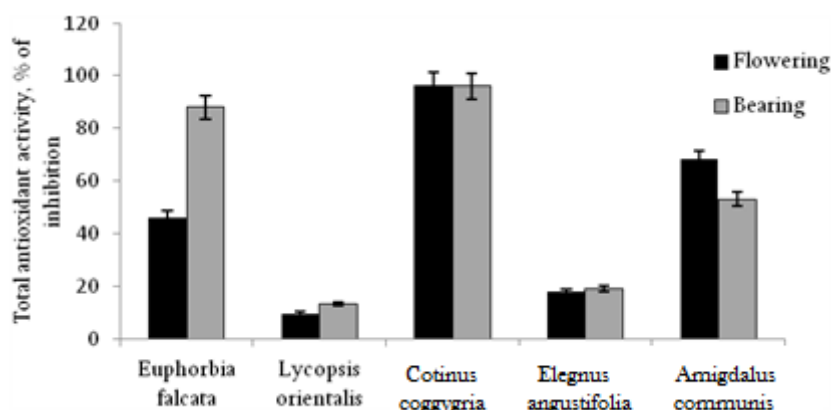


FIGURE 8: Total antioxidant activity of leaves of Iori plateau (East Georgia) drought resistant species in two vegetative (flowering and fruit-bearing) phases

As the studied plants are of medical interest along with drought resistance, the characteristic of total antioxidant activity can serve as a kind of preliminary test in terms of their use as a source of antioxidants. However, this index does not give a complete picture of plants antioxidant capacity and medical value.

According to some authors, an extract with more than 70% of total antioxidant activity is considered as effective; If the index of a sample is 60-70%, it has moderate activity; and data below 60% indicate a weak antioxidant capacity (Luzia and Jorge, 2014). Based on this criterion, smoke tree has demonstrated high antioxidant activity among the tested plants. In sickle spurge this characteristic was weak during the flowering phase and increased in the fruiting phase. The moderate antioxidant activity was revealed in almond, and low – in small bugloss and Russian olive (Fig. 8).

As it was mentioned, this characteristic does not give a complete picture of the antioxidant capacity of the test-objects because it comprises only hydrophilic antioxidants. While among plant antioxidants there are lipophilic compounds as well.

The enhancement of antioxidant activity in the fruit-bearing phase in sickle spurge may be associated with an increase in anthocyanins and phenolic compounds; in small bugloss – with ascorbic acid and anthocyanins. In smoke tree the increase in

ascorbic acid is likely to be offset by a decrease in phenols in the fruit-bearing phase; therefore, the overall level of antioxidant activity has not changed. It is incomprehensible the slight decrease of the antioxidant activity in almond leaves; as its rise on the expense of anthocyanins and phenols enhancement was presumable.

IV. CONCLUSIONS

From the obtained results, it is clear that the studied drought-resistant species have more or less different biochemical stress-adaptive mechanisms. This is not surprising, as such differences may exist not only at species but at the genotypic level within a single species as well (Abuelsoud and Papenbrock, 2019).

For each studied species, it may be identified a stress coping strategy that includes certain enzymatic and non-enzymatic components of the antioxidant system.

Catalase was activated in small bugloss in response to stress enhancement, while from low-molecular-weight antioxidants synthesis of ascorbate-tocopherol and anthocyanins was enhanced. An especially large amount of proline accumulation was noted from osmolytes in its leaves.

Phenols, anthocyanins and proline should be actively involved in drought and high temperature stress in sickle spurge. Russian olive was distinguished by the maximal level of almost all studied indices, compared to other species. Apparently, this is its specific feature. Since some of these characteristics were reduced under increased stress, they still retained their maximal value and obviously played an active role in stress protection of the plant. In particular, high levels of ascorbate, tocopherol and anthocyanins, as well as proline, were found in Russian olive leaves. In addition, the enzymatic antioxidants catalase and peroxidase were activated, and carbohydrates were accumulated.

The protective systems of ascorbic acid and tocopherol, as well as phenolic compounds were active in smoke tree. From osmolytes content of proline increased, while the sugars level was already the highest, compared to all other species. Catalase was activated in response to stress in almonds; protective systems of ascorbate-tocopherol, phenols, and anthocyanins were active from the low-molecular compounds; among osmolytes content of carbohydrates was increased.

The stability of carotenoids protective system of all studied species under stress-enhancing conditions suggests that experimental plants were less sensitive to radiation stress, and water deficiency was the main stress factor for them. All tested species can be recommended as drought resistant, which may be cultivated under water deficiency conditions.

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Physical Properties of Non-Fermented and Fermented Tobacco of Burley Varieties and Lines

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Abstract— *The physical properties of the raw material are an objective indicator of the quality of the tobacco leaves and a reflection of their structure. They are very closely related to the structure and the content of the organic matter in the leaf. The selection of the variety, the applied agricultural techniques, the environmental conditions during the vegetation period, the leaf position, the technological maturity, as well as the conditions during drying, are important factors that have a strong impact on the formation of the physical and technological properties of the tobacco raw material. The tests included 4 varieties and 2 lines, namely: L-8 (control variety, Zimbabwe), Kentucky-22 (USA), B-963 (Bulgaria) and B-1246 (Bulgaria), all in fertile form, as well as the male sterile hybrid lines B-204/15 CMS F₁ and B-206 A/15 CMS F₁. The test was planted in 4 repetitions with a planting density of 90×50 cm according to the Randomized Block System method. After the evaluation of the tobacco according to the current Rulebook for qualitative evaluation of raw tobacco, we separated tobacco material from the middle belt, in order to get an insight into the differences of the physical properties from the examined varieties and lines in the non-fermented and fermented tobacco. The percentage portion of the main (mid) rib, the thickness of the leaves, and the materiality of the leaves, are important physical indicators of the quality of the raw material, and from the obtained results we can point out the line B-206 A/15 CMS F₁, where the average content of the main rib of the non-fermented tobacco leaf is within the limits of 26.82%, in the line B-206 A/15 CMS F₁ there is up to 25.13% portion of the main rib of the fermented leaf. The leaf portion in the newly obtained line is within the range from 73.18% (non-fermented tobacco leaf) to 74.87% (fermented tobacco leaf). The materiality is within the range from 41.26 g/m² for non-fermented tobacco leaf up to 41.90 g/m² for fermented tobacco leaf, and the leaf thickness is 91.5 μm (non-fermented tobacco leaf) up to 77.2 μm (fermented tobacco leaf).*

The obtained data from non-fermented and fermented tobacco, the content of the main rib (%), the thickness of the tobacco leaves (μm), the materiality of the leaves (g/cm²), are determined according to recognized methods that are being applied in the operation of the accredited laboratory - L04 within the Department of Technology, Fermentation and Fabrication at the Scientific Tobacco Institute Prilep.

Keywords— *non-fermented, fermented, tobacco, burley, leaves.*

I. INTRODUCTION

The raw material of the burley type within the composition of the blend cigarettes participates with approximately 30%. In addition to oriental tobacco varieties in North Macedonia, few years ago large-leaf tobacco varieties were grown, there are favorable conditions for these varieties, and therefore we hope that they will be a part of the tobacco production again. The introduction of the burley type in the typical structure of our country was made by Rudolf Gornik. The author (1985) notes that this tobacco type can be successfully grown only on rich soils in and humid climates with frequent rainfall. In the early seventies, this type of tobacco was the point of departure in the search for a variety that will prove to be the best according to most properties (primarily yield and quality). The burley type is dried in the shade in a closed space, it is an integral part of the blend cigarette mixture, and it is used both for a pipe and as a chewing tobacco. The typical characteristic of burley is a leaf with a spongy tissue and a great ability to absorb liquid substances. The content of nicotine and proteins is high, and as a

result of the long drying process, it contains a low amount of sugar, in traces. Its taste is sharp (ammonia-like), it is used for making American blend cigarettes, whereby it participates with approximately 30% in this mixture (Stankovic, 2002, Georgiev, 2002, Radojčić, 2011). The Ministry of Agriculture, Forestry and Water Economy has recognized several burley male sterile varieties of the Scientific Tobacco Institute - Prilep, including: B-96/85 CMS F₁, Burley 1 CMS F₁, B-2/93 CMS F₁ and Pelagonec CMS F₁, all of which are different and have their own specifics. These varieties meet the physical, technological, degustatory and chemical characteristics that are typical of the burley type. Continuously throughout the operation of the Scientific Tobacco Institute Prilep, because there is no ideal variety that is created once and for all, but there is a variety that at one point is better than the others, the Department of Genetics, Selection and Seed Control in the Scientific Tobacco Institute Prilep still continues to create new varieties, as well as many new hybrid lines, including B-204/15 CMS F₁ and B-206 A/15 CMS F₁ which we analyze in the test. This primarily refers to varieties (genotypes) that will provide higher yields, quality and appropriate technological, chemical and degustatory properties. According to Uzunoski (1985), the physical and technological properties of the raw material are an integral part of the quality of the raw material and together with the chemical and degustatory properties they make a natural connection that the quality of the tobacco leaf depends on.

During the tobacco fermentation, oxidation-reduction reactions occur, as well as reactions of hydrolytic decomposition and condensation of specific chemical compounds. These entire reactions make up the complete fermentation process, indicates Boceski (2003). The chemical composition of tobacco changes, through the regrouping of molecules and the decomposition of compounds into end products, which are separated from the tobacco substrate in the air and in the environment. In this way, not only that the quality of tobacco changes, but tobacco also loses weight. This weight loss is caused by compounds that are separated from the organic matter and a part of the hygroscopic water. This loss is a general loss, while the loss of the tobacco's organic mass is a loss of dry mass or fermentation sludge. Some physical changes that occur after fermentation will be seen in the following presentation.

II. MATERIALS AND METHODS

The test was performed within the premises of the Scientific Tobacco Institute Prilep on alluvial-colluvial soil type that was previously prepared and fertilized with 300 kg/ha within the ratio of nitrogen, phosphorus and potassium - NPK 8:22:20. The tests included 4 varieties and 2 lines - creations of the Scientific Tobacco Institute Prilep, as follows: L-8 (control variety, Zimbabwe), Kentucky-22 (USA), B-963 and B-1246 (Bulgaria), all in fertile form, as well as the male-sterile hybrid lines B-204/15 CMS F₁ and B-206 A/15 CMS F₁. The tobacco was planted in 4 repetitions with a planting density of 90×50 cm on June 6, 2018. During the vegetation, the tobacco was dug twice, and nourished with nutrients once with 3-4 g/stalk of 26% CAN (calcium ammonium nitrate). Due to the frequent rainfall, which was sufficient and properly distributed, there was no additional irrigation. During the vegetation of the plant, the same agro-technical measures were applied for all examined tobacco varieties and lines. The harvesting of the tobacco was performed manually, and the drying was performed in special dryers in shade, typical of the burley type. The dry tobacco was estimated according to the applicable Rulebook, and the physical properties: leaf length and width (cm), main rib content (%), tobacco leaf thickness (µm), materiality of the leaves (g/m²), were determined according to recognized methods used by the accredited laboratory - Laboratory-L04 within the Department of Technology, Fermentation and Fabrication at the Scientific Tobacco Institute Prilep (Smokvoski, 2004).

These properties are one of the most significant ones for the cigarette factories because they directly affect the financial effect achieved through the total number of manufactured smoking units of a specific amount of fine-cut tobacco. According to Uzunoski (1985), the technological properties of the raw material are an integral part of the quality of the raw material and together with the chemical and degustatory properties they form a natural connection that the quality of the tobacco leaf depends on.

III. RESULTS AND DISCUSSION

3.1 Main (mid) rib content (%)

The main rib of the tobacco leaf, in addition to its role of a main conducting system for nutrients, has developed into an organ that also has a skeletal function. As a result of the fact that large-leaf tobacco types, one of which is the burley type, have leaves with large dimensions, the percentage portion of the main rib is also higher.

The lateral nervation of the leaf is poorly developed and does not affect the technological processes of the cigarette factories, while the content of the main rib is a very important factor for the manufacturers and processors of large-leaf tobacco varieties. The percentage portion of the main rib in the process of processing directly affects the profitability of the company.

Table 1 and Figure 1 contain data on the portion of the main rib in non-fermented and fermented tobacco of the varieties and newly obtained hybrid lines.

The lowest percentage of the main rib in the examined varieties and lines of non-fermented tobacco, of 26.82%, was found in the line B-206 A/15 CMS F₁, and up to 32.76% in the variety Kentucky-22 which indicates 17.84% higher relative difference than the control variety (L-8). The other varieties and lines of non-fermented tobacco have a portion of 28.20% of the main rib in the line B-204/15 CMS F₁, up to 32.20% in the variety B-963. After the fermentation in all varieties and lines in the test, we observe a lower percentage of the main rib portion, which is a positive indicator in the further processing of the tobacco leaves. The lowest percentage of the main rib portion of the fermented leaves was found in the line B-206 A/15 CMS F₁ (25.13%), whose index compared to its own control of non-fermented leaves is 93.70%. Furthermore, the highest main rib portion of the fermented leaves was found in the variety Kentucky-22 (30.29%), whose index is 92.46%. From the obtained data compared to the own controls of non-fermented leaves, we notice that during the fermentation of the leaves there has been a decrease in the percentage of the main rib portion in all examined varieties and lines.

Hristoski (2013) indicates that the average content of the main rib in the examined varieties by insertions is within the range from 26.82% in insertion X of the control variety, up to 32.16% in insertion B of the variety B-2/93.

Pelivanoska (1999) points out that intensive agricultural techniques increase the percentage content of the main rib by approximately 6% on average compared to the control variety (extensive agricultural techniques).

Risteski (2008), from the three-year research (1999/2001) in six varieties of the burley type (3 domestic and 3 foreign) cultivated in the region of Prilep, in the variant harvested-cured and stalk-cut harvest, came to the conclusion that the content of the main rib is within the range from 27.46% (stalk-cut) in the Chulenec variety up to 32.62% (harvested-cured) in the C-104 variety. Risteski (2006), in his three-year research came to the conclusion that the average content of the main nerve in both variants of harvesting, is within the range from 27.46% to 32.62%. According to Pezović, et. al. (1983), the main rib content of the mid-belt insertions, depending on the production micro-region, is within the range from 19.98% up to 27.78%.

All tested varieties and lines of fermented tobacco show a lower index (non-fermented: fermented leaves). Or the variety B-963 (80.75%) has the lowest index compared to its own control, and the line B-204/15 CMS F₁ (98.94%) has the highest index.

TABLE 1
PORTION OF THE MAIN (MID) RIB OF FERMENTED AND NON-FERMENTED TOBACCO (%)

Variety	Rib Portion in non-fermented tobacco in %	Relative	Rank	Rib Portion in fermented tobacco in %	Relative	Rank	Index	Rank
L-8 Zimbabwe	27.80	100.00	5	26.43	100.00	5	95.07	5
Kentucky-22 USA	32.76	117.84	1	30.29	114.60	1	92.46	3
B-963 Bulgaria	32.20	115.82	2	26.00	98.37	2	80.75	1
B-1246 Bulgaria	28.83	103.71	3	26.56	100.49	4	92.13	2
B-204/15 CMS F ₁	28.20	101.44	4	27.90	105.56	3	98.94	6
B-206 A/15 CMS F ₁	26.82	96.47	6	25.13	95.08	6	93.70	4

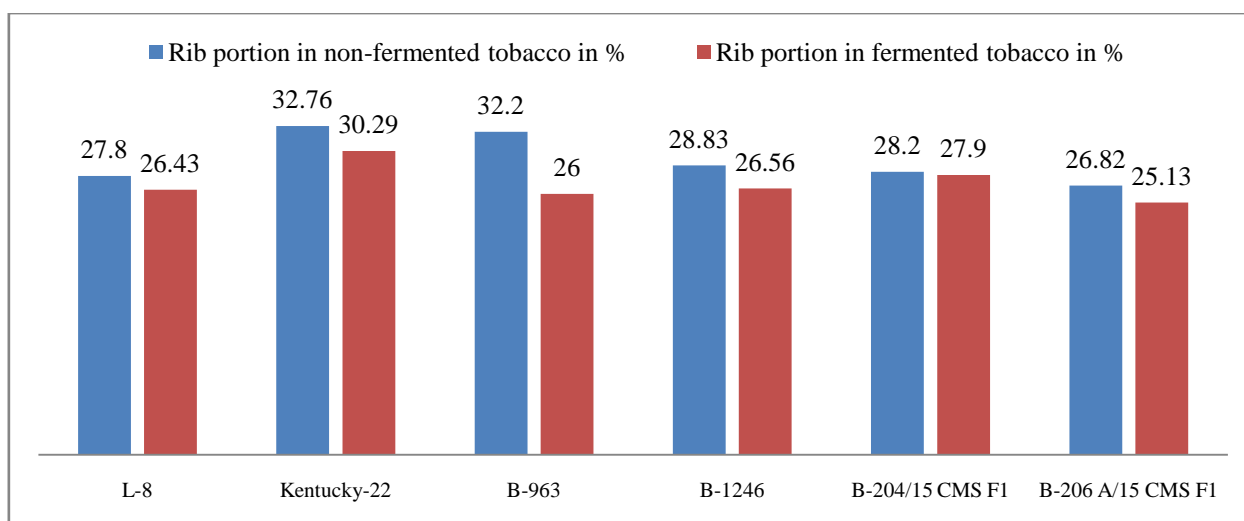


FIGURE 1: Main Rib portion of non-fermented and fermented tobacco in %

According to Uzunoski (1985), in regard to the leaves of the burley type, the share of the main nerve in the total mass of the leaf participates with approximately 30%.

The content of the main rib in the examined varieties is correlated with the cited literary data, and it is within the expectations. We also found that fermented tobacco compared to non-fermented tobacco participates with a lower percentage of the main rib, which depends on the examined variety and line.

3.2 Leaf portion of non-fermented and fermented tobacco (%)

The percentage portion of the leaves compared to the percentage portion of the main rib is inversely proportional, whereby the percentage portion of the leaves is lower in non-fermented tobacco, compared to the fermented leaves where the percentage portion is higher. According to Table 2 and Figure 2, the highest leaf portion of the non-fermented tobacco was found in the line B-206 A/15 CMS F₁ (73.18%), and the lowest percentage was found in the variety Kentucky-22 (67.24%). In the fermented tobacco, the highest percentage portion of the leaf was again found in the line B-206 A/15 CMS F₁ (74.87%), and the lowest percentage portion was found in the variety Kentucky-22 (69.71%). According to the obtained data, all examined varieties and lines of fermented tobacco show a higher index (non-fermented: fermented leaves). More specifically, the variety L-8 has the lowest index (101.90%) compared to its own control variety, and the variety B-963 has the highest index (148.34%). The leaf portion in all examined varieties and lines in fermented tobacco is higher as a result of the reduced percentage share of the main rib, whose index in B-206 A/15 CMS F₁ is 139.07%, in B-204/15 CMS F₁ it is 136.49%, in B-1246 it is 140.26%, and in Kentucky-22 it is 140.91%.

**TABLE 2
LEAF PORTION OF NON-FERMENTED AND FERMENTED TOBACCO (%)**

Variety	Leaf Portion in non-fermented tobacco in %	Relative	Rank	Leaf Portion in fermented tobacco in %	Relative	Rank	Index	Rank
L-8 Zimbabwe	72.20	100.00	2	73.57	100.00	3	101.90	6
Kentucky-22 USA	67.24	93.13	6	69.71	94.75	6	140.91	2
B-963 Bulgaria	67.80	93.91	5	74.00	100.58	2	148.34	1
B-1246 Bulgaria	71.17	98.57	4	73.44	99.82	4	140.26	3
B-204/15 CMS F ₁	71.80	99.45	3	72.10	98.00	5	136.49	5
B-206 A/15 CMS F ₁	73.18	101.36	1	74.87	101.77	1	139.07	4

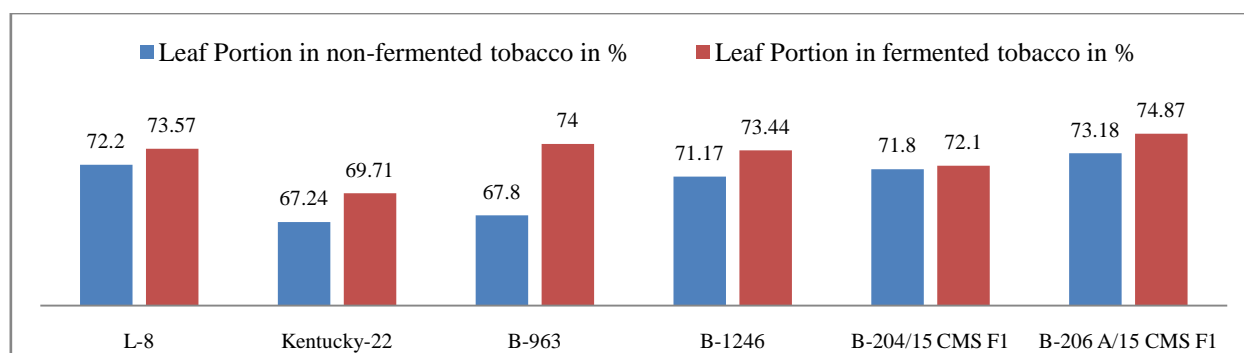


FIGURE 2: Leaf portion of fermented and non-fermented tobacco in %

3.3 Leaf materiality (g/m²)

The materiality or content of the leaf tissue is also one of the important physical properties of the raw material that determine the quality of the leaves. Tobacco leaves that are characterized by roughness, thick or empty leaf tissue, i.e. their materiality is very high or very low, have lower quality i.e. this is a negative indicator of the tobacco quality.

The thinner and more elastic the leaf tissue, the higher the quality. Usually the leaves of the upper belt are slightly thicker and rougher, with a denser cell structure and therefore their materiality is higher. In our tests, leaves from the middle belt were taken. In Table 3 and Graph 3 the materiality of the leaves of non-fermented tobacco is within the range from 35.50 g/m² in the variety B-963, up to 57.08 g/m² in the line B-204/15 CMS F₁. The newly obtained line B-206 A/15 CMS F₁ is characterized by lower materiality (41.26 g/m²) compared to other varieties, which is a positive indicator of the quality of the tobacco raw material. After the fermentation of the tobacco leaves, the materiality is within the range from 53.29 g/m² in the control variety L-8, up to 38.58 g/m² in the variety B-963. From the tests we can reach the conclusion that there is no significant change after the tobacco fermentation, and depending on the variety and the line, the index either increases or decreases by some percentage, and the index (for non-fermented: fermented tobacco leaf) is within the range from 93.17% in the line B-204/15 CMS F₁, up to 108.68% in the variety B-963.

Hristoski (2013) indicates the insertion X in the burley tobacco with an origin from India is characterized by the lowest materiality (33.14 g/m²), and the insertion T with an origin from Greece i.e. Sri Lanka is characterized by the highest materiality (43.70 g/m² and 43.44 g/m²). Pelivanoska (1999) presents data according to which the materiality of the leaves of the burley variety grown in (1996/1998) in conditions of different intensity of agricultural techniques, is within the range from 34.75 g/m² (fertilized and irrigated with 70% of the field water capacity - PWC) up to 52.26 g/m² (fertilized and non-irrigated).

From the studies (1999/2001) conducted with six male sterile varieties of the burley type, Risteski (2008) came to the conclusion that the average materiality of the leaves is within the range from 38.72 g/m² (stalk-cut harvest) in the control variety up to 42.95 g/m² (harvested-cured) in the burley-1 variety.

TABLE 3
MATERIALITY OF FERMENTED AND NON-FERMENTED TOBACCO (g/m²)

Variety	Materiality of the leaf in non-fermented tobacco in g/m ²	Relative	Rank	Materiality of the leaf in fermented tobacco in g/m ²	Relative	Rank	Index	Rank
L-8 Zimbabwe	56.20	100.00	2	53.29	100.00	1	94.82	5
Kentucky-22 USA	47.96	85.34	3	50.30	94.39	3	104.88	2
B-963 Bulgaria	35.50	63.17	6	38.58	72.40	6	108.68	1
B-1246 Bulgaria	44.65	79.45	4	45.07	84.57	4	100.94	4
B-204/15 CMS F ₁	57.08	101.57	1	53.18	99.79	2	93.17	6
B-206 A/15 CMS F ₁	41.26	73.42	5	41.90	78.63	5	101.55	3

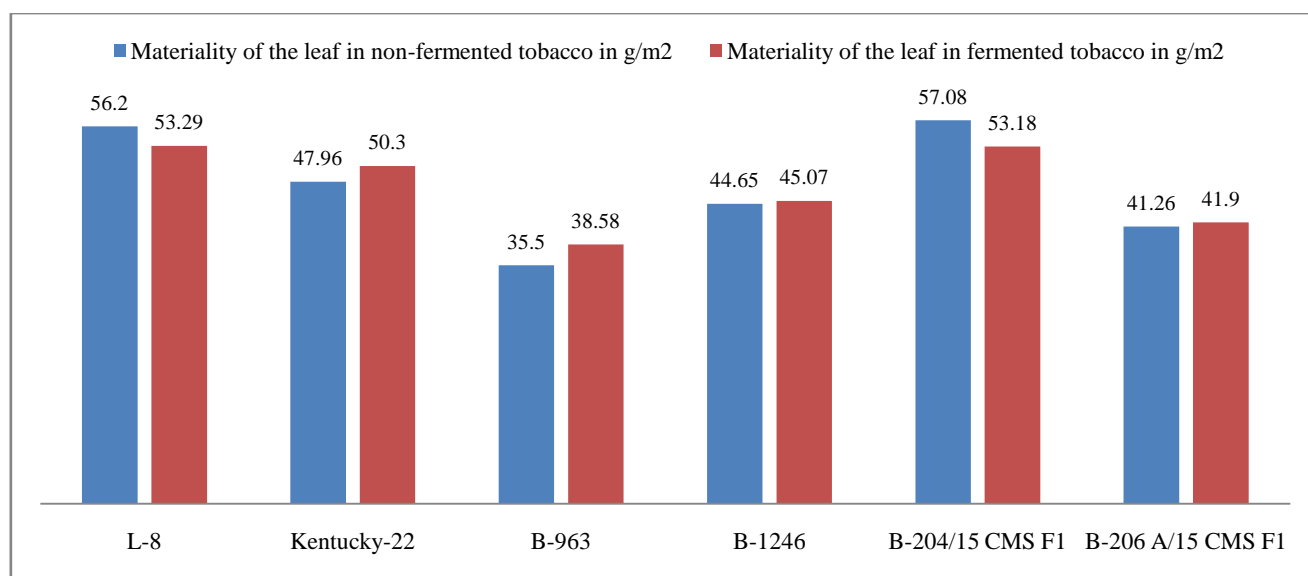


FIGURE 3: Materiality of fermented and non-fermented tobacco in g/m²

3.4 Thickness of the tobacco leaves (μm)

The thickness of the leaf tissue is a reflection of the quality of the raw material. One should bear in mind that in burley tobacco varieties, specifically the lower insertions are characterized by thin leaf tissue. By reducing the thickness of the leaf, the quality of the raw material increases, with the exception of the leaves that are not sufficiently ripe or have become overripe in the course of the vegetation period, or more precisely, empty no-content tobacco. According to Tomič et al. (1977), the thickness of the tobacco leaf varies from 50 to 150 μm . The authors point out that the leaf is thin when the average thickness of the leaf tissue is 70 μm , an average thickness is when the leaf tissue has 70-100 μm and a thick leaf tissue is the one that exceeds 100 μm (micrometers).

The thickness of the leaves depends on the variety, the soil-climatic conditions, the applied agricultural techniques during the vegetation period, the mineral nutrition, the irrigation and the period when the tobacco leaf was harvested.

From the results presented in Table 4 and Figure 4, it is observable that the average leaf tissue thickness in almost all examined varieties and lines is typical for the burley tobacco type, whose tobacco raw material belongs to medium-thick leaves.

TABLE 4
THICKNESS OF NON-FERMENTED AND FERMENTED TOBACCO (μm)

Variety	Leaf thickness in non-fermented tobacco in μm	Relative	Rank	Leaf thickness in fermented tobacco in μm	Relative	Rank	Index	Rank
L-8 Zimbabwe	89.3	100.00	5	78.6	100.00	2	88.02	2
Kentucky-22 USA	95.9	107.39	3	77.3	98.35	4	80.60	5
B-963 Bulgaria	78.3	87.68	6	76.0	96.69	6	97.06	1
B-1246 Bulgaria	102.6	114.89	1	82.5	104.96	1	80.41	6
B-204/15 CMS F ₁	96.3	107.84	2	77.9	99.11	3	80.89	4
B-206 A/15 CMS F ₁	91.5	102.46	4	77.2	98.22	5	84.37	3

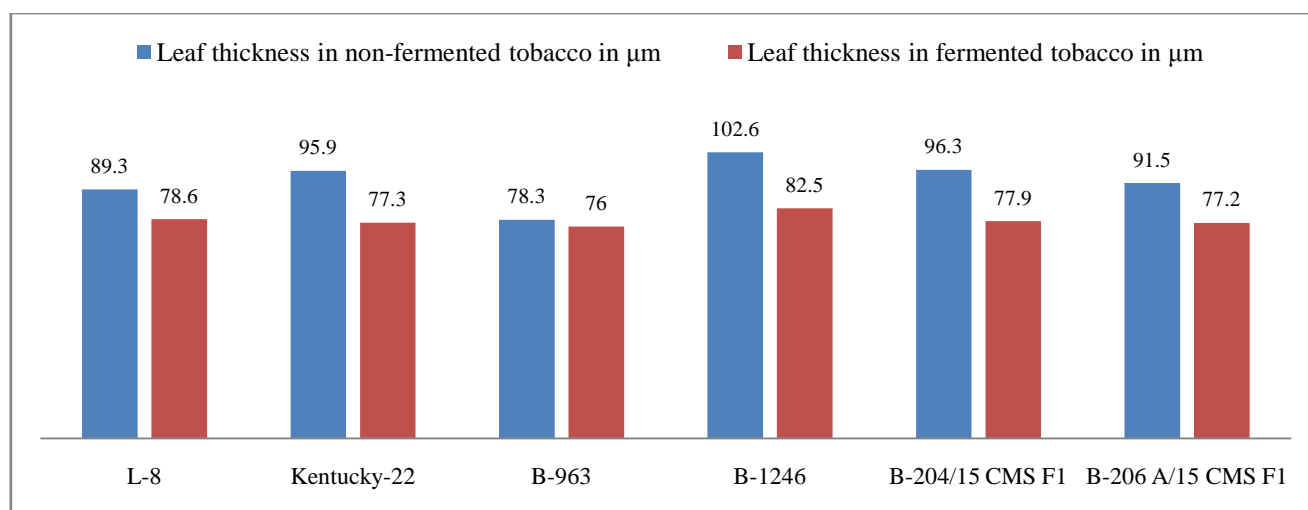


FIGURE 4: Leaf thickness of non-fermented and fermented tobacco in µm

The leaf thickness of non-fermented tobacco of the variety B-1246 (102.6 µm) has the highest value, while the lowest thickness is found in the variety B-963 (78.3 µm). After fermentation, the leaf thickness in all examined varieties and lines decreases, ranging from 82.5 µm in the variety B-1246, up to 77.2 µm in the line B-206 A/15 CMS F₁. The index percentage is lower than the own control in all examined varieties and lines compared (non-fermented: fermented tobacco leaf), it is within the range from 97.06 µm in the variety B-963, to 80.41 µm in the variety B-1246.

Filiposki (1986) concluded that mineral nutrition and irrigation of tobacco have an effect on the reduction of the thickness of the leaf tissue.

Pelivanoska (1999), in her three-year (1996/1998) researches on the impact of agricultural techniques on the quality of the burley raw material, came to the conclusion that the average thickness of the leaf tissue is within the range of 54.00 µm (fertilized and irrigated with 70% of the field water capacity - PWC up to 67.16 µm (control). Risteski (2006), from the researches (1999/2001) in six varieties of the burley type grown in the Prilep region, came to the conclusion that the average thickness of the leaf tissue is within the range of 61.00 µm (harvested-cured) in the variety C-104 up to 71.83 µm (stalk-cut harvest) in the control variety, B-96/85.

The data regarding the thickness of the leaf tissue in the examined varieties from the test are in accordance with the cited literary data, that is, they are within the expectations and are typical of the burley type. The thickness of the leaf tissue is a varietal characteristic and depends on the influence of the soil-climate factors, the applied agro-technique as well as the manner and technology of drying.

IV. CONCLUSION

Based on the obtained results, the following conclusions can be drawn:

- The line B-206 A/15 CMS F₁ is characterized by the lowest percentage portion of the main rib in the examined varieties and lines, where the portion of the main rib in the non-fermented tobacco leaf is 26.82%, and in the fermented one it is 25.13%. The highest percentage of the main rib is of the variety Kentucky-22 in non-fermented tobacco (32.76%) and fermented tobacco (30.29%).
- The leaf percentage portion is highest in the non-fermented tobacco from line B-206 A/15 CMS F₁ (73.18%) and the fermented tobacco (74.87%). The Kentucky-22 variety has the lowest percentage of leaves, the non-fermented has 67.24%, and the fermented tobacco has 69.71% leaves.
- The leaf materiality of the non-fermented tobacco is within the range from 35.50 g/m² in the variety B-963, up to 57.08 g/m² in the line B-204/15 CMS F₁. The newly obtained line B-206 A/15 CMS F₁ is characterized by lower materiality (41.26 g/m²) compared to the other varieties, which is a positive indicator of the quality of the tobacco raw material. After the fermentation of the tobacco leaves, the materiality is within the range from 53.29 g/m² in the control variety L-8, up to 38.58 g/m² in the variety B-963.

- The leaf thickness is highest in the non-fermented tobacco of the variety B-1246 (102.6 μm) and it is 82.5 μm in the fermented tobacco of the same variety compared to other varieties and lines. The lowest leaf thickness is found in the variety B-963 (78.3 μm) in the non-fermented tobacco, and 76.0 μm in the fermented tobacco leaves.

From the tests we can conclude that the newly obtained line B-206 A/15 CMS F₁ has typical physical properties for the burley type.

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Application of Termite Bait with Variation of Methylene Eugenol, Pineapple Peel Extract and Bintaro Liquid Smoke

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Abstract— One of the alternative methods of termite control that can be done is the feeding method. In this study, impregnated bait will be formulated with Pineapple peel extract and Bintaro liquid smoke which functions as a poison for termites. To attract termites to eat poisonous bait, 1% of an attractant compound in the form of methyl eugenol is given to the artificial bait. The concentrations of liquid smoke used were 2 and 4%. Pineapple peel extract is known to have antifeedant activity (reduces appetite) against pests that eat it. The active ingredients in this extract have the ability to damage the digestive mucosa of termites and kill the termite symbiont protozoa so that the ability of termites to digest can be decreased or lost. The concentrations of pineapple peel extract used in this study were 3 and 6%. Based on data obtained from data on time of death, percent mortality and percent palatability of termites to bait, it shows that the best treatment is treatment K7 which causes highest termite mortality for 12 days with palatability of bait is 7.55%.

Keywords— termite, bait, toxic, pineapple peel, bintaro.

I. INTRODUCTION

Coptotermes curvignathus are the main termite types of pests in rubber and oil palm plantations because they can damage the roots and stems of plants and cause the plants to die^[1]. Termites nest in moist soil and wood. West Kalimantan, which is a tropical area with 1.73 million hectares of peatlands and is the fourth largest peat area in Indonesia, has the potential for high intensity of termite attack on its crops^[2]. This is because the soil conditions are wet / moist and contain high organic matter. Termite pest control has been carried out using termiticides by spraying. This technique is less effective in controlling termites, because the level of contact with termites is low and the active ingredients are mostly wasted to non-targets and carried by water / air. In addition, there are negative impacts of the use of synthetic pesticides on the environment and the survival of the ecosystem around the application area.

One of the alternative methods of termite control that can be done is the feeding method. This method is more specific to termites than the spray method and has a higher impact on the termite mortality rate^[3]. The preferred feeding media for termites are materials with a high cellulose content^[4]. Wood with high cellulose and containing methyl eugenol compounds has the highest level of termite palatability. Methyl eugenol is a compound that can attract termites (attractants). This material is mixed with active ingredients that are toxic to termites. This feeding method utilizes the trophallaxis (mutual feeding) properties of termites in one colony^[5]. The worker caste termites whose job is to find food will like and eat the bait, then the toxic active ingredients that have been eaten are spread into the colony and kill the termites.

Based on the nature of these termites, it is expected that the poison bait used has slow action or direct non-lethal toxicity. Termites that carry bait containing poison will carry it into the nest, and the mechanism of trophallaxis and cannibalism between termites can take place. Therefore, a slow action poison was chosen. One of the natural ingredients that has the potential to be used as a slow action poison is the liquid smoke of Bintaro fruit and pineapple peel extract. The liquid smoke of Bintaro fruit contains cerebral compounds which are known to have activities that interfere with termites' nerves or do not directly kill^[6,7]. At its optimum concentration, 7.5% Bintaro liquid smoke can kill as much as 78.42% of termites^[6]. It is also known that pineapple peel extract has antifeedant activity (reduces appetite) against pests that eat it with an optimum concentration of 10%^[8]. In addition, the active ingredients contained in this extract have antimicrobial activity^[9], so that they have the ability to damage the digestive mucosa of termites and kill the termite symbiont protozoa so that the ability of

termites to digest can be decreased or lost. Termites that experience decreased feeding activity will experience hunger and die in their nests and become food for the colony.

II. MATERIALS AND METHODS

Termite harvesting was carried out at a rubber plantation in Sungai Ambawang Village, Kubu Raya Regency, West Kalimantan, Indonesia. The making of bait formulations and application tests was carried out at the Plantation Plant Science Laboratory of Agricultural Technology Department at the Pontianak State Polytechnic.

2.1 Equipment and Material

The equipment used in this study was a set of glass tools, distillator, pyrolysis apparatus, rotary evaporator and test container. The materials used were aquadest, bintaro fruit (*Cerbera manghas*), etanol 96%, methyl eugenol, pineapple peel, sand, and Whattman filter paper no 41.

2.2 Methods

The stages in this research were the pineapple peel extraction, production and purification of Bintaro liquid smoke, phytochemical screening, preparation of termites, bait formulations, application test on termites and analysis of palatability and mortality. The details of the research stages are as follows:

2.2.1 Pineapple Peel Extraction

Extraction of pineapple peel waste using the maceration method. About 10 Kg of pineapple peel is cut into small pieces, then dried for 8 days. The dry sample is then blended dry until smooth. Samples were weighed as much as 3,350 grams and soaked using 96% ethanol solvent with a ratio of 1: 5 (w/v) for 2 x 24 hours and washed 3 times. Filtering is carried out and the filtrate is taken and then evaporated at a temperature of 50°C so that the extract is separated from the solvent.

2.2.2 Production and Purification of Bintaro Liquid Smoke

Twenty Kg Bintaro fruit taken from the field. Then the fruit is washed using clean water and cut into 2 parts using a knife and then dried for 24 hours using a cabinet dryer at a temperature of 60°C until the Bintaro fruit is dry. Then the process of making liquid smoke from Bintaro fruit is carried out using pyrolysis equipment. The burning of Bintaro fruit is carried out in a simple pyrolyzer at 400°C for 8 hours. The liquid smoke obtained is Grade 3 liquid smoke^[6]. Furthermore, distillation is carried out at a temperature of 150°C to purify liquid smoke into Grade 2^[7].

2.2.3 Phytochemical Screening

Phytochemical screening was carried out to determine the content of active compounds in pineapple peel extract and liquid smoke of Bintaro fruit. The phytochemical screening included qualitative tests for alkaloids, flavonoids, tannins, saponins, steroids and terpenoids.

2.2.4 Termites Preparation

Termites are taken from rubber plantation on peatlands located in the Sungai Ambawang Village, Kubu Raya Regency, West Kalimantan, Indonesia. Termites are taken together with the nest and put in a container to be taken to the laboratory.

2.2.5 Bait Formulation

The main ingredient of this stage is filter paper dripped with pineapple peel extract (PPE) and bintaro liquid smoke (BLS) according to the specified treatment with and without methye eugenol (ME). K0= Control negative; K1= 1% ME; K2= 2% BLS+ 1% ME; K3= 4% BLS + 1% ME; K4= 2% BLS; K5= 4% BLS; K6= 3% PPE + 1% ME; K7= 6% PPE+ 1% ME; K8= 3% PPE; and K9= 6% PPE. Then the bait is drained and ready to be applied.

2.2.6 Bait Application Test on Termites

Each of the bait formulations was placed in a container measuring 30 x 10 x 10 cm. Into the container were 33 *C. curvignathus* termites, consisting of 30 worker termites and 3 soldier termites. All containers that have been filled with bait and termites are kept in the dark for two weeks. Each treatment received four replications. During testing the moisture of the sand at the bottom of the test media is maintained by adding water to the sand and the dead termites are immediately removed from the test media. After three weeks the test container was disassembled and the palatability of the bait

formulation and the mortality of *C. curvignathus* were calculated. Each bait formulation was dried on oven for 48 hours at a temperature of 60 ± 2 °C, then weighed.

2.2.7 Lethal Time and Palatability Analysis

Time of death of termite pests was observed at 30 minutes, and every 2 days after bait application for 2 weeks. The faster the time for the pest to die, the higher the rate of poison control in the bait against these pests.

Palatability is based on the weight loss of the bait, which is calculated by the following formula:

$$\text{Palatability (\%)} = \frac{(W1 + W2)}{W1} \times 100\%$$

W1 = bait weight (mg)

W2 = final bait weight (mg)

III. RESULTS AND DISCUSSION

3.1 Phytochemical Screening Analysis

The results of phytochemical screening on pineapple peel extract and Bintaro liquid smoke are shown in Table 1. The results obtained indicate a color change on the addition of reagents. A positive result indicates that the sample contains the phytochemical compounds.

TABLE 1

THE RESULT OF PHYTOCHEMICAL SCREENING OF PINEAPPLE PEEL EXTRACT AND BINTARO LIQUID SMOKE

Phytochemical	Pineapple Peel Extract	Bintaro Liquid Smoke
Alkaloid	+	++
Flavonoid	+	+
Tannin	++	+
Saponin	+	+
Steroid	+	+
Terpenoid	+	+

The table shows that both materials contain all the tested phytochemical compounds. Pineapple peel extract contains more tannin which was observed from a very concentrated color change. The presence of tannin compounds in a material is known to act as an antimicrobial because it is lipophilic^[9]. Tannins can inhibit damage to microbial cell walls, causing leakage and cell death. In termite bait, the presence of tannins in the bait formulation is expected to be toxic to the termite symbiont protozoa, thereby disrupting the termite's metabolism in digesting their food. In the liquid smoke of Bintaro fruit, it can be observed that it contains more alkaloid compounds. This is shown from the very concentrated color change in the test results. Alkaloid compounds are known to be neurotoxin, so that termite death due to these compounds can be observed from the symptoms of changes in termite movements^[7].

3.2 Lethal Time Analysis

Time of termite mortality can be observed in Figure 1. In all treatments, the number of termites that died increased with time. The 24 hours after application, there is no dead termites were seen. In general, death occurred on the second day. K7 was the treatment with the highest number of mortality and increased until the twelfth day. In some treatments, it appears that the pest mortality rate is quite slow (seen from the sloping graph) as in the K3 and K4 treatments, while in other treatments it shows that the pest mortality rate increases significantly from day to day.

Based on the data analysis, it shows that K3 and K4 treatments are significantly different from other treatments. The treatment with the highest percentage of mortality is K5 (4% Bintaro liquid smoke), was not significantly different from treatment K7 (6% pineapple extract + eugenol), but significantly different from other treatments. This shows that giving pineapple extract and liquid smoke at higher concentrations can kill pests more and more quickly.

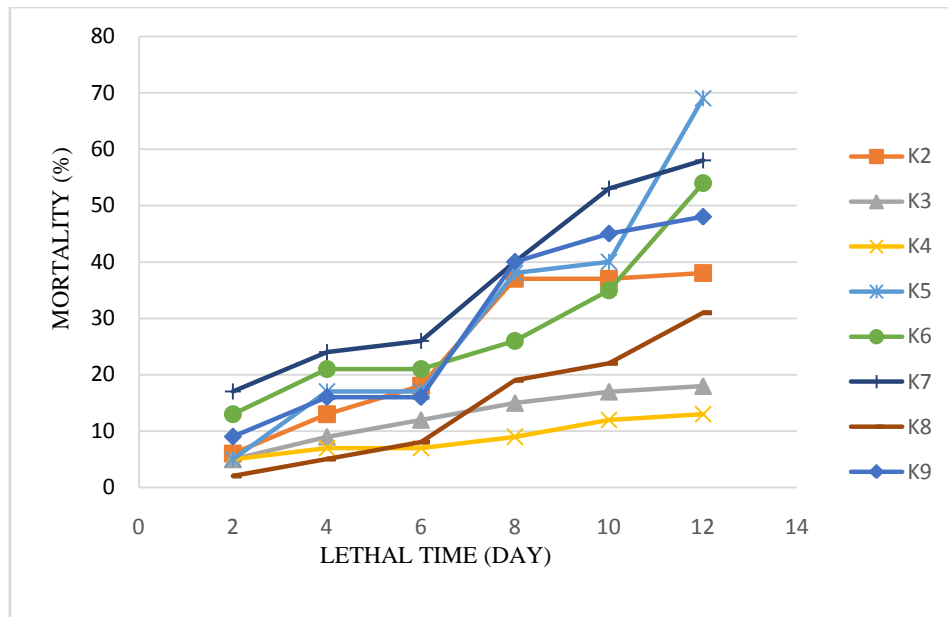


FIGURE 1: Time of Termite Mortality

3.3 Palatability Analysis

Based on the percentage palatability in Figure 2, it can be seen that the application of pineapple peel extract has a high percent palatability compared to giving Bintaro liquid smoke to the bait. This can be caused by the smell of the liquid smoke which termites do not like.

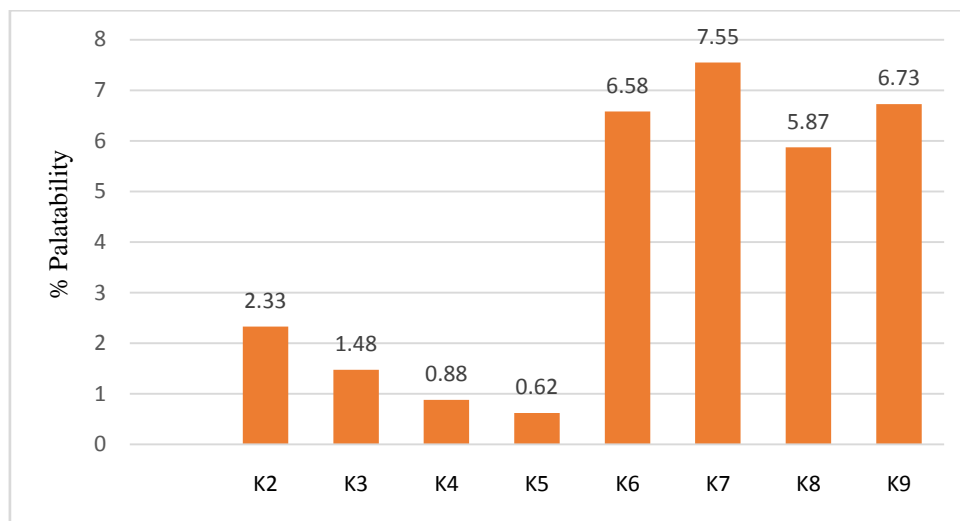


FIGURE 2: The Number of Termite Palatability for Bait Formulations

Pest mortality can be affected by high levels of termite feeding on the toxic bait given. The more bait is eaten, the higher the active compound in the formula is in the target pest's body, so that the poisoning reaction will be faster and the mortality will be higher. However, this was not the case in this study. It can be seen in Figure 2 that the K5 treatment is the treatment with the lowest percent palatability, meaning that the interest in eating termites to bait is very low. This can also be caused by the strong smell of liquid smoke. As much as 0.62% of the bait is eaten by termites, but the termite mortality rate in this treatment is the highest, followed by K7. The comparison between uneaten and eaten bait is shown in Figure 3. The author suspects that the pest's mortality in K5 treatment occurred due to pests of hunger due to no food consumed for days. Different things can be seen in the treatment of K6, K7, K8 and K9. It can be seen that the percent palatability of termites to bait is quite high, ranging from 5.87 to 7.55%. This is also directly proportional to the high rate of pest mortality, so it can be concluded that the death of pests in this treatment is due to the reaction of active compounds in the feed formulations given. In addition, it can be seen that in treatment K6 and K7 have a higher percent palatability than K8 and K9. This shows that the presence of eugenol in the bait increases the interest of termites to the artificial bait compared to bait without eugenol.

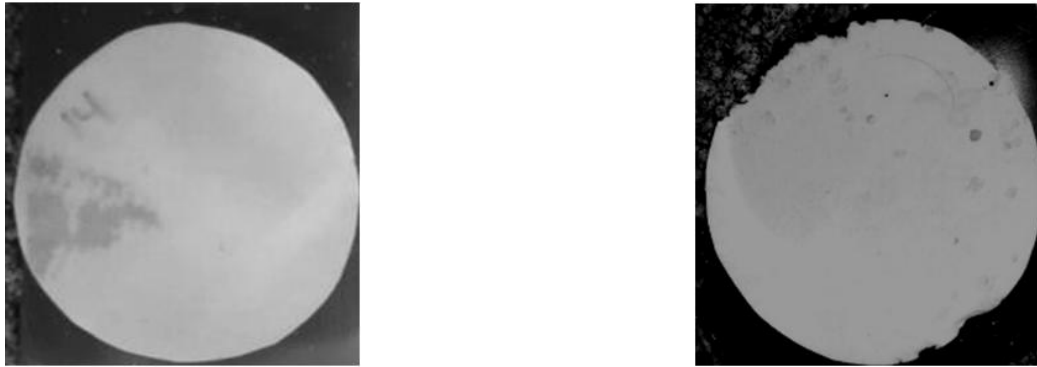


FIGURE 3: Uneaten Bait (Left) and Eaten Bait (Right)

IV. CONCLUSION

Treatment K7 (pineapple peel extract 6% + Eugenol 1%) is the best treatment, where termites have the highest feeding interest in this bait formulation and a highest mortality for 12 days of bait application. Pineapple peel extract formulations with and without the addition of eugenol given to high cellulose sources have the potential as a slow toxic attractant for termite control in rubber, oil palm and several other types of plants on peatlands. This is shown from the data on mortality and percent palatability after application.

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The Political Economy of Agricultural Development in Northern Nigeria

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Abstract— *The paper critically examines the political economy of agricultural development in Northern Nigeria. The agriculture resource has been a significant sector in the Nigerian economy in the past decades, and is still a key sector regardless of the oil boom; principally it provides employment opportunities for the teeming population, eradicates poverty and contributes momentarily to the growth of the economy. The Agricultural sector suffered neglect during the hey-days of the oil boom in the 1970s. However, sustained economic development cannot be achieved without economic growth. Consequently, economic growth is necessary for sustained economic development. In the same vein, given the enormous resource endowment both in human capital and natural resources available in Nigeria, the performance of the Northern Nigeria economy has been far below expectation. Consequently, the contributions of agriculture to economic growth can be examined through the roles of the sector in the economy. The most direct contribution of agriculture to economic growth is to increase in incomes of smallholder farmers and therefore their purchasing power. The economic growth in Nigeria depends to a large extent on growth in the agriculture sector. However, the article reveals some factors that negatively impacting agricultural development in Nigeria include land tenure systems; increasing populations and constantly decreasing farmland size; inadequate of capital particularly for the adoption of improved agricultural technology; never-ending conflicts in the Northern Nigeria; throng rural-urban migration; low level of education; systemic corruption of government officials; excessive dependence of oil economy to the exclusion of agricultural economy; unfavourable economic development policies; inadequate infrastructure among others.*

Keywords— *Political, Economy, Northern Nigeria, Oil boom, Agriculture, Development.*

I. INTRODUCTION

The Northern Region Nigeria is a region that contradicts its natural endowments. In spite of the existence of several economic resources such as tin, kaolin, a variety of agricultural products and a huge fertile land, the people remain in abject poverty leading to plethora of crisis in forms of insurgency, electoral violence and crime (Titus, *et al.* 2017). Out of the six geopolitical zones in Nigeria, three are in the northern part of the country and they have the worst indices of poverty compared to the other zones. The Northern Nigeria, occupying 70% of Nigeria's land mass, with its huge solid mineral deposits, growing mining industry. The Northern region has 50 million Muslims, one of the largest Muslim populations in Africa. These Muslims are of Hausa/Fulani ethnicity, which is the largest ethnic subgroup in Africa and Nigeria.

The Northwest with 77.7% North-central having 67.5% and Northeast with 76.3% (United Nation, 2012). Northern Nigeria becomes a hub of joblessness, crime, illiteracy, maternal mortality, early marriage and recently, farmer herdsman crisis. The political economy of agriculture in Northern Nigeria, this has been developed over the past four decades and has been important not only in the study of agriculture in Sub Saharan Africa but more widely. Nigeria has long been a case of interest for the study of political and economic development. The political economy of the Nigerian society has suffered pitfalls resulting in an economy powered by visionless leaders, known for reckless spending, over-invoicing, diverting state finances into private account, thereby plunging the country into economic, social and political wretchedness (Ganiyu, *et al.* 2014). It is

a common knowledge to many across the globe that the Northern region performs at a level of productivity far less than its full potential and other part of the country. This is further exacerbated by population increase which outpaces food production per capita the latter of which is in decline; this situation also results in the concomitant effect of increased importation of food in the region (Nwachukwu, 2016).

Nigeria is a great example of a post-colonial developing nation and its developmental history encompasses very important lessons on the political and economic obstruction of the developing world (Ugwuanyi, 2014). The country as a colonial entity enjoyed thriving and boom in the agricultural production and the mining of mineral resources such as iron ore, tin and coal. The Nigeria foreign exchange was earned from the aforementioned resources. Each region had a proportional advantage through which it made its significant contributions to the centre/federal government. The Northern Nigeria for instance, was known for groundnut production, the West for her cocoa while the East produced palm oil. During the British colonial era, the politics of development was already manifested even with the construction of railway line through those areas that had products that could yield some revenue to the centre (Osita-Njoku, 2016).

According to Njoku (1998), the British political economy in Nigeria was along the line of economic exploitation of the colonized by foisting it into the path of the European capitalist economic system. Ezeanyika (2010) argued that the overall subordination of colonized nations by dominating foreign power is to “keep the colonized people in complete political subjection, and to maximize local human and natural resources”. However, there are some factors that negatively impacting agricultural development in Nigeria include land tenure systems; increasing populations and constantly decreasing farmland size; inadequate of capital particularly for the adoption of improved agricultural technology; never-ending conflicts in the Northern Nigeria; through rural-urban migration; low level of education; systemic corruption of government officials; excessive dependence of oil economy to the exclusion of agricultural economy; unfavourable economic development policies; inadequate infrastructure among others.

In October 1, 1960, Nigeria gained her political independence from the British colonial master, the production of crude oil had changed the dynamics of the Nigerian political economy. Oil boom production is now the mainstay of the economy. Luqman and Lawal (2011) stated that hardly could anything be written about the political economy of Nigeria without reference to its history of oil production. Despite the fact that the oil industry remains the mono-economic fulcrum of the Nigerian economy, its contribution to economic development and improvement of the living standards and welfare of Nigerians remain doubtful. This was because Nigerians case is simply a situation of poverty amidst plenty. Instead of our visionless leaders reinvesting the resources from excess crude oil sale into development of infrastructure like power, education, health, public transportation, water, housing, good roads and national security, among other sectors, what the country witnessed is a political class deeply entrenched in corruption and siphoning oil income abroad, whereas country like Indonesia turned crude oil income into productive investment for the benefits of the citizen (Luqman and Lawal, 2011; Ganiyu, *et al.* 2014). This unpleasant situation can not augur well in the development of a stable political and economic institution necessary for building a strong and successful nation.

II. THE POLITICAL ECONOMY

The political economy of agriculture has long been a puzzle and the concept is not new in sociological and political discourse. Political economy can be understood as the art or study of the management of a country in the sense of macro or public household but taking into account political, economic, social, cultural, institutional and other factors that come into play and not forgetting the complex interactions between them (Swinnen 2010; Nwachukwu 2016). The term emerged as a distinct field of study and was developed in the 18th century as the study of the economies of states, or politics (Groonwegen 2008). In the present day, political economy, where it is not used as a synonym for economics, may refer to very different things including Marxian analysis, applied public choice approaches emanating from the Chicago School and the Virginia school, or simply the advice given by economists to government or public on general economic policy or on specific proposals (Groonwegen 2008).

However, political economy is applied here as the study of the social relations, particularly the power relations, that mutually constitute and impact the production, distribution and consumption of resources in agricultural development. According to Maier (2008), political economy approach interrogates economic doctrines to disclose their sociological and political premises. Basically, the term is refers to as economic ideas and behaviour, not as frameworks for analysis, but as believes and actions that must themselves be explained. Consequently, it deals with the interactions between economic policies and their social and political context. Eboh (1999) maintained that political economy is an approach-cum-subject concentrating on the structure of markets and government, the incentives, abilities and behaviour of economic agents, policy makers, civil

servants and society at large. Fundamentally, political economy provides a more critical understanding of the foundation upon which all social and political life is built. Hence, offering a more distinct analysis than that offered by mainstream approaches that are patently failing a majority of people at the behest of a minority (Aregbeshola, 2011).

III. POST-COLONIAL SITUATION OF AGRICULTURAL DEVELOPMENT IN NIGERIA

Nigeria became independent nation in 1960. At this point, it was the expectation of the citizens that the country will turn around for good. The nation was led to believe that following independence in Nigeria in 1960, the nation would continue to progress into greater magnitude of prosperity that would usher in quality standards of living for the citizens. This believe was not just a mere wishful thinking simply because of the much foreign exchange earned through agricultural exports and both cash and food crops were massively produced. It was very convenient for the world to perceive Nigeria to be the future giant of Africa (Osita-Njoku, 2016). The citizen strongly believed that indigenous leaders who have taken over the mantle of leadership had an answer to the way forward from where the British colonial masters left the country. Indeed, the different regions of the country experienced financial explosion till the late 1960's from engaging in the agricultural production and export of agricultural produce they cultivated under colonialism (Iwuagwu, 2008). At that time, through concerted effort of the organisation of land and labour by peasant farmers, Nigeria was one of the world's major producers, not only of palm oil, but also of cocoa and groundnuts, as well as cash crops for domestic consumption.

However, in 1970's the country experienced the abandonment of the aforementioned agricultural resources with all attention focused on oil income which now made the crude oil the mainstay of the Nigeria economy, that is, a mono-product economy (Dodo, 2009; Osita-Njoku, 2016). As a result, all the developmental programmes initiated at different points in time by different administrations could not be implemented to achieve anticipated results. At this juncture, a close attention will be given to the developmental programme implemented after independence; their level of success and the reasons for their failure. Post-colonial development strategies in Nigeria were articulated under the various national development plans namely, the first National Development Plan (1962-1968); second National Development Plan (1970-1974); the third National Development Plan (1975-1980); the fourth National Development Plan (1985-1990).

The main objective of Nigeria's National Development Plan was to preserve and possibly, to surpass the average rate of growth of 4% per year of its gross domestic product at constant prices. To achieve the aim, government planned annually investment of approximately 15% of Nigeria's gross national product. Given that agriculture was the major strength of Nigeria's economy, and which was largely identified with the rural areas, policy attention and governmental investment in it were seen as direct and indirect avenues of developing the rural areas.

3.1 First National Development Plan Period (1962-1968)

The first National Development Plan was strategically put in place. The Plan succeeded at first raising the rate of economic growth, and to increase control of the economy. Looking at the first plan critically it was simply a continuation of the British colonial development policy that placed emphasis on transportation and communication, such as facilitating the movement of raw materials out and finished product into the country (Ibietan and Ekhosuehi, 2013). As a result, instead of moving the nation forward in her developmental effort in terms of achieving the main objectives for which it was set up, the nation experienced some structural inconsistencies (Dodo, 2009; Ugwuanyi, 2014). The plan itself did not articulate any clear statement or policy on rural infrastructural development. Instead, emphasis was placed on encouraging the assemblage of agricultural produce for export purpose, without strengthening the real agricultural base of the country by providing necessary infrastructures such as good road network, electricity, agricultural processing facilities, and potable water, among several others.

3.2 Second National Development Plan (1970-1974)

The second National Development Plan (1970-1974) came as a post-civil war development ingenuities. It was also during this plan period that Nigeria had the 'phenomenon of oil resource boom'. Principally, the plan was aimed at: a) building a united, strong and self-reliant nation; b) building a great and dynamic economy; c) building a just and egalitarian society; d) building a land of bright and full opportunities and; e) building a free and democratic society (Marcellus, 2009; Ikeanyi, 2009). The plan placed high priority on reducing the level of inequality among the social classes and between urban and rural areas. Basically, one important feature of the second National Development Plan as perceived by Marcellus (2009) was its democratic content, having emerged from a participatory process that involved stakeholders at every level of governance. According to Leonard (2006), the discovery of oil in the 1970's the nation's economy has been a mono-economy because of the over dependence on the oil sector, which provides 95% of foreign exchange earnings, and about 80% of budgetary

revenues. He further stated that, the oil boom, as it has killed other resources of revenue for the country. 'Oil boom' soon translated into struggle for 'oil rents' which led to massive corruption at every levels of governance (Oyefusi, 2007; Akinyetun, 2016). Huge spending and import of food characterized the state activity while agriculture that served as the mainstay of the economy was relegated to the background. Given the consistent poor funding of agriculture with rural development in Nigeria, government massive dependence on oil revenue during this period meant that all policies on rural development could no longer be on the agenda of government. Moreover, oil boom has been key to Nigeria's political economy since 1970's, giving rise to syndrome called the 'resource curse'. This includes a revenue monoculture, endemic corruption, political uncertainty, communal tension and heightened conflict (Akpan, 2012).

3.3 Third National Development Plan (1975 – 1980)

The third National Development Plan (1975-1980), rural development was reconsidered based on government egocentric conviction that such investment will make substantial contribution in closing the yawning gap between the demand for food and the supply capacity of the home-based industries. Consequently, government developed interest in modernizing agriculture and introducing original initiatives to strengthen the agricultural and food base of the nation (Lewis, 1977). Even though the objectives of the plan looked similar to those of the second national development plan, there was a considerable and comprehensive approach as the plan give emphasis to the need to lessen regional disparities with the intention of promotes national unity through the adoption of unified rural development. Increased budgetary allocations were provided to fund diverse and interrelated rural development sectors as the provision for nationwide agricultural programmes (Ugwuanyi, 2014). However, during the third national period, some agricultural development programs were initiated include:

- Operation Feed the Nation (1976).
- River Basin and Rural Development Authorities (1976).
- Agricultural Development Project (ADP) which was funded by the World Bank.
- Green Revolution Programme (1980).

It is imperative to note that from the first to the third national development plans, there was observable progressive budgetary improvement to boost agricultural productivity. Olorunfemi and Adesina (1998) observed that there were increasing financial allocation for agricultural development; and the third national development plan had the highest allocation for agricultural development. However, such inconsistent development interest was not enough for transforming rural communities without corresponding investment in rural infrastructures such as motorable roads, electricity, health care, pipe borne water, among several others (Leonard, 2006).

3.4 The Fourth National Development Plan (1981 – 1985)

The Fourth National Development Plan (1981-1985): This was a civilian government development plan which emphasized among other things the need for balanced development of the different sectors of the economy and of the various geographical areas of the country. Unfortunately, the fourth development plan period was threatened by fall in oil revenues and equally delays in agricultural modernization due to decline in funds in-flow and consequently an increase in the quest for imported foods (Eneh, 2008). The plan laid emphases on the need for rural infrastructural development as a means of increasing the standard of living in the rural communities. As a result, the following allocations were made:

- ₦924 million was released by Federal Government for eleven River Basin Development Authorities towards construction of boreholes, dams, feeder roads and jetties.
- Federal and State Government's allocation of ₦645 million and ₦700.4 million respectively for electrification purposes.
- For rural water supply schemes, ₦2, 805 million was allocated while the local governments in some states allocated a total of ₦311, 824 million for water projects (Olayiwola and Adeleye, 2005).

Many local governments and various states governments stated numerous policy issues that could improve the standard of living of the rural dwellers. However, the Fourth national development plan was characterized by huge debt servicing which resulted from various foreign loans obtained in the previous years; increased import bills in the midst of a drastic fall in crude oil export revenue (Iheanacho, 2014). However, it is imperative to note that the overthrow of Nigeria's second civilian administration, the Second Republic headed by President Shehu Shagari, at the end of 1983 and of the military government

of General Muhammadu Buhari in 1985 brought to an end the fourth development plan (Eneh, 2008; Ikeanyibe, 2009; Lawal and Abe, 2011).

IV. WHY NIGERIA DEVELOPMENT PLAN FAIL

Although Nigerian developments plans have assisted in moving her forward positively change, however the plans still have various defects:

4.1 Misplacement of Priorities

Like the colonial ones, the policies of the post-independent plans also demonstrated a basic lack of urgency. For instance, the iron and steel industry that was in the first and second development plans and was initiated for projected completion during the fourth plan. This characterized a lag of twenty years; however this project was repeatedly acclaimed the cornerstone of Nigeria's industrialization (Ejumudo, 2013). Another good example closely related to the aforementioned includes petrochemicals, fertilizers, the petroleum refinery, liquefied petroleum gas and other heavy industries.

4.2 Systems Corruption

Nigeria is a country where corruption has been institutionalized and raised to the level of a structural parameter. As a matter of fact, corruption has become part of the value-system of a society, a condition par excellence. It could be stressed that from the strategic corruption of the cement armada to the mega tonic corruption of the second Republic the nation have consciously or unconsciously, created systemic corruption in every sphere of the sector and the trend has continued through the third and fourth republic (Ejumudo, 2013).

4.3 Absence of Relevant Data

Planning relies essentially on accurate data. It is unfortunately however to note that accurate data is a very scarce commodity in Nigeria due to problems arising from the inadequacies of the federal office of statistics, the disinclination of Nigerians to reveal information and the outright manipulation of data for financial or other gains. Furthermore, due to the challenge of relevant data since independence, government has not been able to as answer the simple question "How many are we in Nigeria?" Unfortunately, a nation that does not know its population would certainly not be in position to determine the other important statistics essential for strategic planning (Ejumudo, 2013).

4.4 Flaws in the Strategies adopted by the Government, particularly in Rural Infrastructural Development Programmes

The fusion of government activities reveals that since independence, several developmental programmes have been initiated the government to combat rural infrastructural problems, which are far from solving the problem due to flaws in the strategies adopted by the government including; (1) using only the development plans as an instruments for programming resource allocations for different sectors of the economy hence failing to recognise the facts that various rural communities having different ecological situations in Nigeria, differ in the nature and degree of their needs (Okafor, 1985); (ii) duplication of programmes between the different tiers of government (Olayiwola and Adeleye, 2005).

4.5 Inadequate Executive Capacity

This is one of the greatest problems of development plans in Nigeria in the area of implementation. In reality, it is frustrating to plan the execution of programmes which require the availability of organization, institutions and skills which the economy does not possess and cannot normally be expected to generate during the plan period. Executive capacity also encompasses the existence of knowledgeable contracting firms and basic socio-economic infrastructure including competent hands to run the civil service and allied government machinery (Ejumudo, 2013).

V. BOKO HARAM INSURGENCY AND AGRICULTURAL DEVELOPMENT IN NORTHERN NIGERIA

Since 2009 Nigeria has been in the grip of a violent Islamic insurgency by the extremist sect widely known as Boko Haram. More than 25,000 people have been killed in Boko Haram attacks across Northern Nigeria with thousand displaced. The insurgency has negatively impacted on agricultural productivity and distribution networks from the north to the southern part of the country and vice versa (Adebisi, *et al.*, 2016; Kah, 2017, Adelaja, *et al.*, 2019).

Boko Haram literally means "*Western Education is divinely forbidden*" and therefore should not be allowed to prevail among nations; particularly, Muslim dominated states. This evil group has attacked and destroys churches, mosques, schools, police stations and private and public owned facilities. In fact, Boko Haram is the most dangerous insurgent group in Nigeria

which has led to the displacement of smallholder farmers in Northeast of Nigeria and subsequently affects the agricultural development of the region. This insurgency has made many farmers relocate to other places where there is peace in order to save their lives, causing them to leave their farmlands behind as it is not mobile (Adetiloye, 2014; Mustapha, 2015). The activities of Boko Haram insurgency and the Fulani herdsmen has negatively influence agricultural development in the Northeast of Nigeria and its environs. Honestly, the destructive effect of Boko Haram insurgency in the North East Nigeria continues to be a source of worry to all and sundry.

The menace caused by Boko Haram insurgency in the North has been a great threat to the Nigeria business environment as farming and other business activities are being harmed. Nomadic cattle herders who have been taking their cattle out for grazing have abandoned their businesses, as the environment is no longer safe for them. The lives of those that are still involved in the agri-business are also at stake, because an attack of Boko Haram can occur at any moment. This has led to decreases in milk production, meat production and by extension an increase in the price of the cattle (Kah, 2017). Poultry farmers in the region also do not find their business lucrative anymore, as the people purchasing the goods have been displaced. As a result, this has led to poor transportation, high transport cost, displacement of properties and high risk to lives (Babagana, *et al.*, 2018).

The evil activities of Boko Haram are not only felt in Nigeria but in some other parts of the Africa. Particularly, the neighbouring countries such as Cameroon, Chad, Niger Republic other countries sharing border with Nigeria in the North East part are also affected. Traders from these countries can no longer come to Maiduguri (Nigeria) to buy or sell. The border was closed some time ago, restricting importation or exportation from the countries which affected their economy (Adebisi, *et al.*, 2017).

VI. CONCLUSION

Insecurity is one of the major impediments to agricultural development in Northern Nigeria. The local economies in the North were especially disrupted by the surge in insecurity allegedly cause by Fulani herdsmen, banditry and Boko Haram insurgency. In fact, Nigeria has been consistently ranks as one of the most unsafe country in the world. The current security crisis and political instability already posed serious challenges to agriculture and disrupt major economic activities in the region thereby making Northern Nigeria a geographic disadvantage. The impact of Boko Haram and Banditry on the Nigerian economy is localized for now, but the instability has had an effect on agricultural products from the North and has severely reduced cross-border trade with neighboring countries. Insecurity in Nigeria has four main effects on agricultural development and investments along the agricultural value chains:

- 1) Reduced human mobility;
- 2) Reduced access to inputs and markets;
- 3) Increased theft of various assets;
- 4) Increased prices of inputs and products.

The effects of insecurity on the Northern agricultural sector are largely due to the risk of being attacked by insurgents and banditry.

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Influence of Presowing Irradiation and High Concentrations of Salts on Wheat

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Abstract— To investigate the salt resistance of Georgian endemic wheat species – Makha (*Triticum aestivum* subsp. *macha* (Dekapr. & Menabde) McKey) and Zanduri (*Triticum timopheevii* subsp. *zhukovskiyi* (Menabde & Ericzjan) L. B. Cai) on the one hand, and the effect of pre-sowing irradiation on growth and development of the same species, on the other, experiments with 1.5% solutions of NaCl and Na₂SO₄ and pre-sowing treatment with ultraviolet irradiation (UV) (C section of the ultraviolet, distance 30cm from the source, irradiation for 1h) have been carried out. Wheat species were affected with stressors separately and in combination. The percentage of seed germination and length of 5-6 week seedlings was studied. Obtained results demonstrate that:

1. Zanduri seeds are equally resistant to both chloride and sulfate salinization, while Makha seeds revealed more resistance to chloride salinization.
2. Irradiation of Zanduri seeds with C section of UV-radiation inhibited germination, while treatment with the same spectrum of Makha seeds in contrary, led to significant activation of the process. This effect of seeds pre-sowing irradiation was maintained during the growth and development stages as well.
3. Pre-sowing UV treatment of Makha seeds has canceled the inhibitory effect of NaCl on seed germination; while in variants with pre-sowing UV treatment and further processing with Na₂SO₄ and Na₂SO₄+NaCl even the stimulation of seed germination was mentioned.
4. In order to increase the seed resistance to chloride and sulfate salinity, we consider it advisable to irradiate Makha seeds with UV before sowing; however, the optimal dose of radiation should be selected.

Keywords— chloride salinity, sulphate salinity, ultraviolet irradiation, wheat.

I. INTRODUCTION

Under natural conditions plants usually experience impact of several unfavorable factors. That is why the resistance to the simultaneous exposure to several stressors is of great importance in selection. Moreover, studies have shown that a plant's response to the combined effect of several stressors differs from the response to a single stress exposure (Craufurd and Peacock, 1993; Jiang and Huang, 2001; Pnueli, *et al.*, 2002); e.g. it was established that activation of diverse genes took place in response to different stresses in *Arabidopsis* the (Mittler *et al.* 2004). Moreover, literary data prove that between the responses to different stresses synergic or antagonistic relations may exist (Walter, 1989; Sandermann, 2004).

More common are data on the simultaneous effects of drought and high temperature stresses on agricultural crops (Craufurd and Peacock, 1993; Jiang and Huang 2001). It has been established that the combined effects of these two stressors have a far more detrimental effect on plant growth and development than the effect of each individually (Jagtap *et al.*, 1998; Wang and Huang, 2004).

Natural ultraviolet (UV) radiation and soil salinization are the stressors that often cause problems for agricultural crops.

Soil salinization is one of the most acute environmental problems in today's agriculture. According to the Food and Agriculture Organization of the United Nations (FAO), more than 20% of the world's sown area and half of its irrigated land is salinized due to secondary salinization and alkalination and became unsuitable for agriculture. Over the next 25 years, 30%

of irrigated land is expected to be salinized, and by 2050 this number will increase to 50% (Gogue, 2014). Georgia is no exception in terms of soil salinization - the total area of saline soils is 1.6% (Urushadze and Blum, 2014).

One of the approaches to solve this problem is identification of existing salt resistant species, or breeding of new, resistant forms.

Investigation of the effect of UV irradiation on plants is very popular as well. B and C sections of UV radiation are known to affect negatively plant growth, photosynthesis, and other vital processes, due to the production of active forms of oxygen. It is well known that these forms of oxygen damage cell macromolecules and can even cause its death (Toncheva-Panova *et al.*, 2010; Zu *et al.*, 2010; Schreiner *et al.*, 2012). However, on the other hand, it is established that low doses of UV-B or UV-C may enhance stress adaptive responses in the plant, due to activation of enzymatic and non-enzymatic defense systems (Lavola *et al.*, 2003; Katerova and Todorova, 2011).

To study the effect of UV radiation on plants is interesting from another point of view, as well. In particular, today there is a growing interest in pre-sowing treatment of seeds by physical, environmentally safe methods. Numerous data are available on the positive influence of pre-sowing treatment of seeds with various types of electromagnetic radiation, including UV, on both yield quantity and quality, as well as on plant protection against various diseases (Dubrov, 1963; Ghallab and Omar, 1998; Delibaltova and Ivanova, 2006).

Investigation of the effect of high concentrations of salts and pre-sowing treatment with UV on economically important crop - wheat was the aim of the study. It included several aspects:

1. Testing of the salt resistance of experimental wheat species;
2. Investigation of the impact of direct UV irradiation on the growth and development of the studied species;
3. Studying of the effect of seeds pre-sowing irradiation on the salt resistance of plants, developed from these seeds.

According to the purpose of the study, the impact of each stress factor on research objects was studied separately and in combination. The results obtained allow judging how pre-sowing irradiation affects the salt resistance of the studied wheat species.

II. MATERIALS AND METHODS

2.1 Experimental plants

Georgian endemic species of wheat Makha - *Triticum aestivum* subsp. *macha* (Dekapr. & Menabde) McKey and Zanduri - *Triticum timopheevii* subsp. *zhukovskiyi* (Menabde & Ericzjan) L. B. Cai were selected as test objects. According to recent classification they are regarded as sub-species. Experiments were made on a widely spread species of mild wheat - *Tr. aestivum* L. as well. Experimental seeds were kindly provided by the department of plant genetic resources of the Institute of Botany of Ilia State University (Georgia).

Selection of Makha and Zanduri for experiments was stipulated by the data on their salt resistance (Badridze *et al.*, 2009). Mild wheat was taken for comparison, as widely spread dominant, as well as relatively salt resistant culture (Wang and Xia, 2018).

Experiments were performed using solutions of sodium chloride (NaCl) and Glauber' salt (Na₂SO₄). The salts solutions were prepared from chemically pure Chimex LTD reagents. An ultraviolet bactericidal lamp ZW40S 19W-21199 (Biobase bioindustry (Shandong) Co. LTD), that emits 253.7 nm waves (corresponding to UV-C section), was used for seed irradiation.

2.2 Description of the experiment

At the first phase of the experiment, observations were made on seeds sown on Petri dishes. Experimental seeds were immersed in a solution of potassium permanganate for several minutes, for disinfection (the part of the seeds which were irradiated before sowing did not need disinfection), then washed under running water and dried on a filter paper.

Part of the tested seeds before sowing, was irradiated with UV bulb (ZW40S 19W-21199 Biobase bioindustry (Shandong) Co. LTD) for 1 hour. A distance of the irradiated material from the bulb - 30 cm. Prepared for sowing seeds was placed on Petri dishes in the amount of 50 pieces per one dish. According to the objectives of the work, 8 experimental variants were provided:

1. **Control, without any impact:** seeds were placed on filter paper soaked in distilled water.

2. **Impact with sodium chloride:** seeds were placed on filter paper soaked in 1.5% solution of sodium chloride.
3. **Impact with Glauber's salt:** seeds were placed on filter paper soaked in 1.5% solution of Glauber's salt.
4. **Joint exposure of seeds with table and Glauber' salts:** seeds were sown on filter paper soaked in a solution of both salt mixtures at a total concentration of 1.5%.
5. **Seed UV treatment:** irradiated seeds were placed on filter paper soaked in distilled water.
6. **Combined exposure to UV irradiation and sodium chloride:** irradiated seeds were sown on filter paper soaked in 1.5% sodium chloride solution
7. **Combined exposure to UV irradiation and Glauber's salt:** irradiated seeds were sown on filter paper soaked in 1.5% solution of Glauber's salt
8. **Joint exposure with irradiation and both salt solutions:** irradiated seeds were sown on filter paper soaked in 1.5% solutions of both salts.

Petri dishes with experimental seeds were placed at room temperature (24°-26°C) under natural day/night illumination. The filter paper of the test-variants periodically was wetted with distilled water to keep the paper from drying out and to maintain the concentration of active salts at the same level. The germination of seeds on Petri dishes and further growth and development of sprouts up to the three-leaf phase was observed. The growth and development of seeds was assessed by Mano's 9-point scale (Badridze *et al.*, 2009).

Two weeks after sowing, the germination percentage of seeds (germinated seeds amount/number of total seeds× 100%) was calculated (Fig. 1).

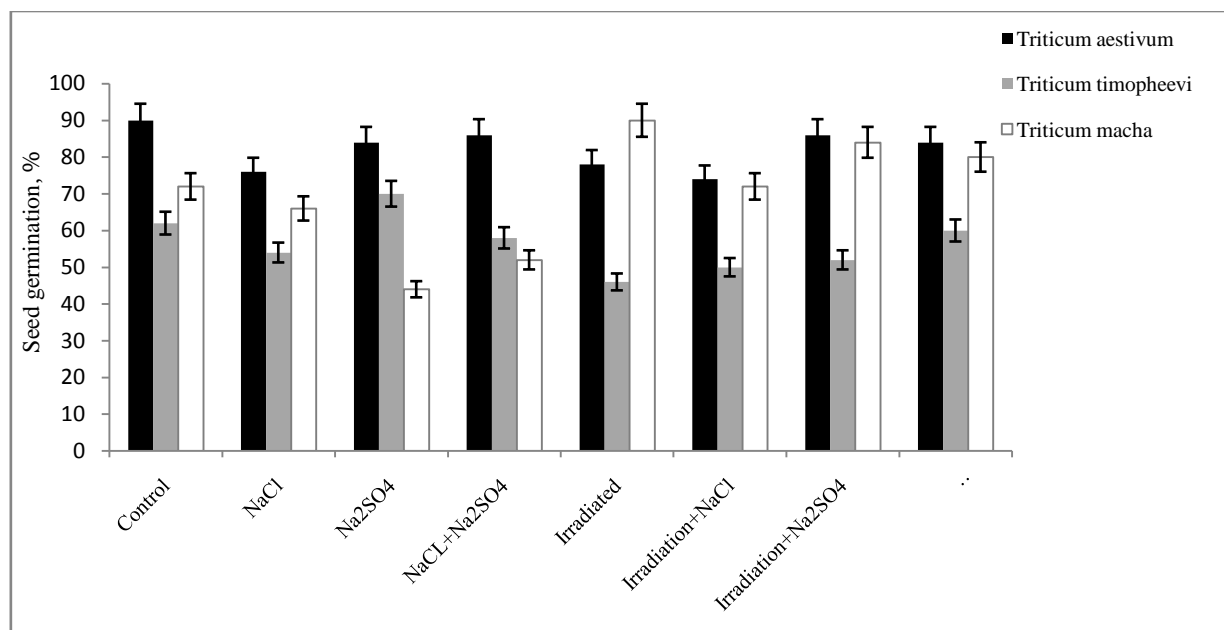


FIGURE 1: Germination percent of soft wheat (*Tr. aestivum*), Zanduri (*Tr. timopheevi*) and Makha (*Tr. macha*) seeds under the influence of sodium chloride (NaCl), Glauber's salt (Na₂SO₄) and UV-C irradiation

At the second step of experiments the three-leaf phase sprouts were transferred in soil, in plastic containers. Experimental plants were watered with distillate as needed. At this stage of the experiment observation of plants growth and development was continued. The height of plants was measured periodically. The data taken in stem elongation phase, in the 5th-6th weeks of development were used for calculations. The experiment was stopped in stem elongation phase when plants had at least 5 leaves.

2.3 Statistical processing of data

One way ANOVA and Tukey's multiple comparison tests were used to test differences between the means. All calculations were performed using statistical software Sigma Plot 12.5.

III. RESULTS AND DISCUSSION

3.1 Percent of germination

Seed germination is one of the major and important stages in the life cycle of a plant, on which the crop is highly dependent. It has been established that soil salinization has negative impact on seed germination of many plants, including crops. Moreover, this effect is multilateral. In particular, seed swelling is inhibited, due to the low osmotic potential of the environment; the activity of many enzymes is changed, as well as hormonal balance, nutrient supply is inhibited, etc. (Parihar *et al.*, 2015).

The stressful effect of salinization is mainly attributed to sodium ions. A number of papers have shown that the coexistence of anions may increase the toxicity of Na^+ . Moreover, in many species sulfate salinity has more toxic effect than chloride (Renault *et al.*, 2001; Martin *et al.*, 2015). However, according to some authors, chloride salinity in some cases is more toxic than sulfate (Munns *et al.*, 2002).

Two weeks after sowing, the number of germinated seeds which were likely to continue development was evident. That is why the percentage of seed germination was calculated at this stage of development (Fig. 1).

From the obtained results it is clear that the exposure to sodium chloride reduced germination rate of Zanduri seeds (by 8%, $p < 0.05$) compared to the control; while Glauber's salt, in contrary had a stimulating effect - the index increased by 8% ($p < 0.05$).

The degree of Makha seeds germination under the influence of salts, especially Glauber's salt, reduced, compared to the control (by 6% in the case of sodium chloride application, by 28% in the Glauber salt variant, $p < 0.05$) (Fig. 1).

The percentage of seed germination under the influence of salts solutions was reduced in soft wheat as well. However, in contrast to Makha, this species was found to be more sensitive to sodium chloride (the emergence rate decreased by 14% when exposed to sodium chloride and by 6% when exposed to Glauber's salt, $p < 0.05$) (Fig. 1).

Seed germination percentage decreased in the variant of combined application of both salts (compared to the control). The combined action of both salts on Zanduri seeds partially preserved the effect of sodium chloride, i.e. the studied index was slightly lower than the control (by 4%, $p < 0.05$) (Fig. 1).

In the case of Makha, the result was averaged between the sodium chloride and Glauber's salt data; while in soft wheat it was closer to the result obtained in the variant with Glauber's salt (Fig. 1).

Thus, at early stages of germination and growth, chloride salinity had more negative effect on both Zanduri and soft wheat than sulfate one; In Makha, the result was opposite. In addition, an antagonistic relationship between the anions of the used salts was found, which was evident in the variant of exposure to both salts. In the case of Macha, this should be due to the attenuation of the inhibitory effect of sulphate anions in the mixed solution of both salts, compared to the Glauber salt variant; In the case of soft wheat, sulfate anions seemed to "remove" the negative effect of chloride ions and the result was similar to the data obtained with sulfate anion (Fig. 1,2).

Seed irradiation means that the dormant seed is supplied with excess energy which is stressful for it. Free radicals (active forms of oxygen) generated in seeds during irradiation, as well as change in the conductivity and electrical potential of the seed and cell membrane are likely to be the starting point for the activation of various metabolic systems, including the antioxidant one. This, in turn, should be reflected on plant further development and formation of adaptation mechanisms to unfavorable environmental conditions (Dubrov, 1963).

From the obtained results, it is clear that the used dose of UV radiation had a depressing effect on the germination of Zanduri seeds and reduced it by 16% ($p < 0.05$) compared to the control variant. The inhibitory effect of irradiation on seed germination was maintained in the salt exposure variants, albeit to a lesser degree: the germination of Zanduri irradiated seeds under the influence of sodium chloride reduced by 12% ($p < 0.05$), compared to the control; in the case of Glauber's salt - by 10%. In the case of combined action of both salts the inhibitory effect of irradiation was eliminated by the stimulating effect of sulfate-anion on the seed germination ability and the result was closer to control (Fig. 1).

Irradiation had a positive effect on Makha seeds germination ability and increased it by 18%, compared to the control variant ($p < 0.05$) (Fig. 1). Seeds UV irradiation seemed to eliminate the negative effect of salts in Makha: the percentage of seed germination in "irradiation+salt" variants was either equal to the control (when exposed to sodium chloride) or exceeded it

(when exposed to Glauber's salt). The stimulating effect of irradiation was maintained in both salts exposure variant as well (Fig. 1).

As for the mild wheat, presowing UV irradiation diminished its seeds germination by 12% ($p < 0.05$). The inhibitory effect of irradiation did not increase in "irradiation+salt" variants and results were similar to those of salts impact variants ($p < 0.05$).

Different abiotic stresses (drought, radiation, salinization, high or low temperatures, heavy metals, etc.) usually affect the plant in combination. The plant has several molecular mechanisms to cope with stress. These are general or specific mechanisms of resistance that are complex and depend on the strength and duration of stress; as well as the tissue or organ on which it acts (Vahdati and Leslie, 2013).

In the conducted experiments, tested species of wheat were exposed to two stressors - high concentrations of salts and UV radiation. Thus, the impact of stressors would lead to the "switch-on" of appropriate protection mechanisms in the plant, which may be discussed by the comparison of the results of experimental and control variants.

Obtained results demonstrate that the applied dose of UV radiation was a stronger stress for the experimental species than the high concentration of salts (especially this is said for Zanduri). However, irradiation of seeds in experimental species induced various reactions. In Makha it presumably activated the general mechanisms of protection against stress, which were also directed against salinity. This may explain the partial neutralization of the negative effects of salts by irradiation.

Opposite - inhibitory effect of pre-sowing irradiation on seed germination was revealed in soft wheat. Presumably, irradiation and exposure to salts resulted in the activation of the same stress protection mechanism in this species; thus, no amplification of the negative effect occurred in the joint exposure variants of the two stressors (Fig. 1).

Moreover, at the seed germination stage of experimental plants, an antagonistic relationship between the seeds pre-sowing irradiation and exposure to salts was found. This was especially evident in the exposure variants of both salts: the inhibitory impact of radiation on seed germination in both Zanduri and soft wheat was overleped by salts effect (irradiation+salt variants), manifested in a reduction of radiation effect. Presumably, the protective response to salinity stress in the early stages of development was faster than the response to radiation. As this anti-stress reaction should be directed generally against stress, abatement of the inhibitory effect of radiation in "radiation + salts" variants was evident.

3.2 Height of seedlings

Growth inhibition is one of the manifestations of the negative effect of salinity stress on plant. This is due to two reasons: a plant has difficulty of absorbing water from the soil due to the low water potential in the latter, on the one hand. Moreover, after salts penetrate into the transpiration current, they cause leaves damaging. It has been established that sodium ions negatively affect the development of leaves more than roots. They accumulate in leaves and cause necrosis of aged leaves, which starts from the tip and edge and spreads throughout the total leaf (Tester and Davenport, 2003; Parihar *et al.*, 2015). Thus, the height of the shoot, which is related to the growth and development of the leaves, provides some information about the effect of stressors on the plant.

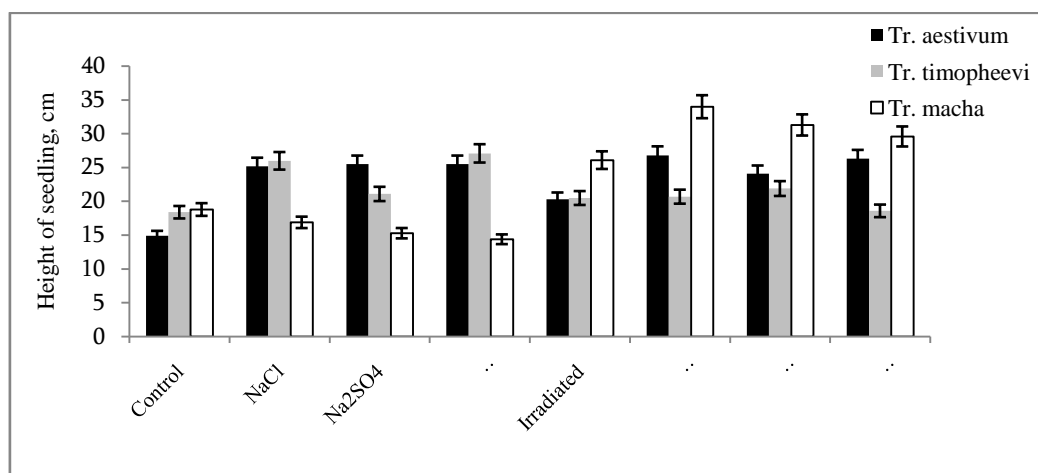


FIGURE 2: Influence of seeds treatment with sodium chloride (NaCl), Glauber's salt (Na₂SO₄) and UV-C irradiation on the height of seedlings of soft wheat (*Tr. aestivum*), Zanduri (*Tr. timopheevi*) and Makha (*Tr. macha*)

Comparison of heights of 5-6 week experimental seedlings cleared, that the negative effect of exposure to 1.5% sodium chloride solution, which was revealed at the germination stage in Zanduri and soft wheat, no longer was evident at seedling's growth phase, and even stimulation of growth was mentioned, compared to control ($p < 0.05$) (Fig. 2). The stimulating effect was also observed in Glauber' and both salts-exposure variants (compared to the control, $p < 0.05$). The reason for such results probably was the transplantation of seedlings into the soil. Since the experimental plants were transferred into the soil, they were irrigated with tap water. Evidently, the high concentration of salts which affected the plants till the three-leaf phase (before being transferred in the soil) were washed out and diluted into the soil. The plants no longer suffered from the inhibitory impact of high concentrations of salts, and the stress removal stimulated their growth. Such an effect may be the result of the activation of stress protection systems at the germination phase, which was caused by the salt stress and retained even after its removal.

In the case of Makha, the inhibitory effect of salts on the growth of 5-6 weeks old seedlings was well expressed, and completely eliminated in the "irradiation + salt" exposure variants. It may be assumed that UV radiation caused such a strong stimulation of stress protection systems in Makha seeds that this effect prolonged in the plant for a long time after germination and led to the complete cancellation of the inhibitory effects of applied salts (Fig. 2). It should be noted, however, that the combined effect of radiation and salt was stronger than only radiation.

The inhibitory effect of UV irradiation, which was observed in Zanduri during the seed germination phase, was no longer observed in the stem elongation phase and the results were statistically close to the control variant ($p < 0.05$). The negative effect of irradiation was also "removed" in combined exposure variants of irradiation and salts ("irradiation+NaCl" and "irradiation+NaCl+Na₂SO₄" variants. $p < 0.05$), and the results were statistically similar to the control variant, or even a slight growth stimulation was mentioned ("radiation+Na₂SO₄", $p < 0.05$), compared to the control (Fig. 2). Presumably, the used dose of UV radiation was found to be more stressful for Zanduri than the applied concentrations of salts. As it was mentioned, the protective response to salinity stress in early stages of development was faster than to radiation, although it was generally directed against stress. Therefore, in the seed germination stage in "irradiation +salts" exposure variants the inhibitory effect of irradiation abated. Later irradiation in Zanduri also led to the activation of certain defense systems. As UV radiation appeared to be the strongest stress for Zanduri wheat, the activity of radiation defence system surpassed the salt protection system. This may explain the cancellation of the growth-stimulating effect of the salts exposure variants (non-irradiated) in the "radiation+salt" exposure variants (Fig. 2).

IV. CONCLUSIONS

1. Zanduri seeds are equally resistant to both chloride and sulfate salinization. However, in terms of resistance to chloride salinity, it even surpasses soft wheat. Makha seeds are more resistant to chloride salinity than sulfate.
2. Irradiation of Zanduri seeds with UV-C radiation caused inhibition of germination, whereas irradiation with the same spectrum of Makha seeds on the contrary led to significant activation of germination quality. This effect of radiation was maintained during the growth and development stage.
3. The inhibitory effect of sodium chloride on the germination of Makha seed was cancelled by pre-sowing UV treatment; while pre-sowing UV irradiation of seeds with further application of Glauber's salt, or joint exposure to Glauber's and sodium chlorides even had a stimulating effect on their germination ability, compared to control.
4. In order to increase the seed resistance to chloride and sulphate salinities, we consider it advisable to irradiate Makha seeds before sowing; however, the optimal dose of radiation should be selected.

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Enriched Mesquite Piperidine Alkaloid Extract Improves the Performance in Growing Goats

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Abstract— This study aimed to evaluate levels of enriched mesquite piperidine alkaloid extract (MPA) comparison with sodium monensin on the nutrition and growth performance of goats fed diets with high concentrate content. Thirty Anglo-Nubian crossbred goats, 120 days of age, and initial body weight 21.82 ± 0.11 kg were distributed to the following diets: 0 (no additive), with MPA 9.2, 18.4, and 27.6 mg kg⁻¹ or monensin (MON) 2.7 mg kg⁻¹. The diets with MPA did not differ ($P > 0.10$) from the MON diet for the intake and digestibility of DM and OM. However, NDF_{ap} and CP intake (g kg⁻¹ BW^{0.75}), MON showed a higher mean compared to MPA, and their digestibility coefficients did not differ. There was a linear increase ($P < 0.05$) for the intake and digestibility of CP and NFC with the MPA levels. The metabolizable energy (ME) and daily weight gain (DWG) presented a quadratic effect ($P < 0.05$) with peaks estimated at 17.4 and 14.8. There was no difference ($P > 0.10$) for microbial nitrogen synthesis, and microbial efficiency decreased linearly ($P < 0.05$) with the MPA levels, but MPA did not differ ($P > 0.05$) from the MON. Nitrogen retention (NR, g day⁻¹) increased ($P < 0.05$) with the MPA levels due to the linear increase of N intake (NI) and digested nitrogen (DN). For the diet with 27.6 mg kg⁻¹ MPA, the DWG decrease occurred due to the lower digestible energy intake and microbial protein synthesis efficiency.

Keywords— Growth Promoter, Performance, Phytogetic Additive, Prosopis Juliflora, Rumen Fermentation.

I. INTRODUCTION

The efficiency of energy and protein utilization in ruminants can be improved by manipulating the rumen microbial population or by the digestion process [1-2]. Ionophore antibiotics are classic supplements in diets showing proven effectiveness in increasing feeding efficiency and reducing the incidence of disorders digestive in diets with high concentrate content [3-4]. For goats maintained in confinement, dose of monensin at 20 mg kg⁻¹ in the feed, recommended by manufacturers, is indicated only for the prevention and control of coccidiosis and the dose of 11 mg kg⁻¹ of feed for three weeks or 8 mg kg⁻¹ of body weight for five days showed no signs of hepatotoxicity [5].

However, many countries have banned the use of antibiotics as growth promoters as a food safety precaution against the occurrence of antibiotic residues in the carcasses [6-7]. In this sense, the use of new technologies is being investigated based on the premise of public health and sustainably increased productivity.

Vegetable secondary compounds extracts, when included in the diet, can affect a wide range of rumen microorganisms [8, 4, 9, 10]. *In vitro* studies carried out with extracts from *Prosopis juliflora* (Sw.) DC (mesquite) pods or leaves have demonstrated the bactericidal potential and antimethanogenic [11-12]. Also, bioactive actions as fungicide, nematicide, and cytotoxic [13-24]. These properties were attributed to the presence of piperidine alkaloids with the greatest pharmacological importance, such as juliprosopine and juliprosine [12].

Mesquite piperidine alkaloids (MPA) are amphoteric molecules of character basic, acting as blockers of Ca^{2+} channels (proteins binding calcium, CaBPs) or as inhibitors of acetylcholinesterase (AChE) in eels and tick [13, 25]. The first action can inhibit sensitive strains to antibiotics similar to ionophores [26]. The binding of piperidine alkaloids to these membrane CaBPs could affect the transport and storage of Ca^{2+} in the bacterial cell, reducing its survival [27]. The second action is a consequence of pH-dependent interactions that prosopine, juliprosopine, and juliprosinine had at the active site of the AChE, mainly relating to hydrogen bonds and cation- π interactions [13].

The MPA showed positive effects on *in vitro* rumen fermentation products and improved the performance without affecting health in sheep fed diets with levels ranging from 2.3 to 31.5 mg kg^{-1} DM [11-12, 28-29]. Because of the scarcity of information with goats, the present study aimed to evaluate the effect of MPA levels (0; 9.2; 18.4; 27.6 mg kg^{-1}) compared to sodium monensin in diets with high concentrate content for growing goats on nutrient intake, digestibility, growth performance, rumen microbial synthesis, and nitrogen balance.

II. MATERIALS AND METHODS

2.1 Ethical Considerations, management, treatments and diets

All the animal care and handling procedures were approved by the Ethics Committee on Animal Use of the State University of Southwest Bahia (UESB), with protocol number 23/2013.

This experiment was conducted in the goat's farming sector of UESB, Itapetinga Campus, State of Bahia, Brazil. Thirty Anglo-Nubian crossbred goats, 120 days of age, and initial body weight 21.82 ± 0.11 kg were housed in individual (1.5×1.0 m) stalls equipped with feeders and drinkers and provided free access to water, feed *ad libitum*.

Animals were distributed in a completely randomized experimental design, with five diets: Diet 1: sodium monensin (MON) 2.7 mg kg^{-1} DM; Diet 2: No additives; Diet 3: enriched mesquite piperidine alkaloid extract (MPA) 9.2 mg kg^{-1} DM; Diet 4: MPA 18.4 mg kg^{-1} DM and Diet 5: MPA 27.6 mg kg^{-1} DM and six animals/treatment. The acclimation period lasted 14 days for adaptation to roughage: concentrate ratio of 20:80 and more 14 days for adaptation to experimental diets. The experimental period was 89 days divided into three sub-periods for sample collection over 5 days. The total dose of monensin was approximately 278 mg kg^{-1} of feed DM per 103 days.

TABLE 1
INGREDIENTS OF THE DIETS AND CHEMICAL COMPOSITION OF TIFTON 85 HAY (BERMUDA GRASS) AND CONCENTRATE

Item	(g kg^{-1} DM)	
Tifton 85 hay	205.3	
Milled corn	202.8	
Soybean meal	571.8	
Mineral salt ¹	20.6	
TDN ²	755.0	
ME (MJ kg^{-1} DM) ³	11.8	
Chemical composition (g kg^{-1} DM)	Tifton 85 hay	Concentrate
Dry matter (g kg^{-1} NM)	830.5	879.1
Organic matter	944.4	951.1
Crude protein	97.9	189.3
Neutral detergent fiber corrected for ash and protein	761.3	208.9
Acid detergent insoluble protein	498.5	154.7
Ether extract	31.9	34.1
Non-fiber carbohydrates	36.5	496.9
Hemicellulose	346.2	177.7
Cellulose	356.6	66.1
Lignin	168.6	11.7

¹ Mineral salt, composition 120 g Ca/kg, 87 g P/kg, 147 g Na/kg, 18 g S/kg, 590 mg Cu/kg, 40 mg Co/kg, 20 mg Cr/kg, 1.8 g Fe/kg, 80 mg I/kg, 1.3 g Mn/kg, 15 mg. Se/kg, 3.8 g Zn/kg, 300 mg Mo/kg, 870 mg F/kg (max.), 95% solubility of phosphorus (P) in citric acid at 2% (min.).

² Total digestible nutrients and ³Metabolizable energy were calculated on tabulated values for individual feed ingredients according NRC (2007).

The diets were formulated following the recommendations of the National Research Council [30] to meet the nutritional requirements of growing goats with an estimated weight gain of 180 g day⁻¹. The diets were supplied twice a day, at 0600 h and 1600 h, with a ratio of 20% Tifton 85 hay and 80% concentrate (total mixed ration, TMR). Tifton-85 hay was used as roughage and chopped to 5.0 mm. Samples of the ingredients and formulated diets were collected and examined for analysis of their chemical compositions (Table 1). The concentrate portion of the diet was a combination of milled corn, soybean meal, and mineral mixture.

2.2 Obtaining enriched mesquite piperidine alkaloid extract

Mature pods of *Prosopis juliflora* (SW) D.C. were obtained from located in the Brumado municipality, Bahia State, being manually harvested from June to July 2017. The pods were sun-dried for three days consecutive and then processed in a Wiley knife mill (A. H. Thomas, Philadelphia, PA, USA) with a 1-mm sieve. The macerate was then percolated and the extracted solution was concentrated in a vacuum evaporator (rotary Fisatom Evaporator – model 802; São Paulo, Brazil) at -600 mmHg and a controlled temperature of 45°C for obtaining the crude ethanol extract (CEE). The CEE was partitioned using acid-base solutions and organic solvents according to the methodology of [31].

Part of the CEE (100 g) was subsequently solubilized in 1.6 M acetic acid aqueous solution (AcOH, 200 ml) and the resulting solution was filtered to obtain acidic aqueous solution I (AAS-I). The AAS-I was extracted with chloroform (CHCl₃) in two successive 150 ml washes, thereby obtaining acidic aqueous solution II (AAS-II). The AAS-II was alkalized with sodium hydroxide (NaOH) until pH 9.0 and called basic aqueous solution I (BAS-I). The BAS-I was triple-washed with 100 ml of CHCl₃, obtaining basic aqueous solution II (BAS-II). The BAS-II was subjected to double washing with sodium chloride solution (NaCl), resulting in basic aqueous solution III (BAS-III) which was subsequently dehydrated with 5 g of sodium sulfate (Na₂SO₄), homogenized, and allowed to stand for 2 hours.

Next, the BAS-III containing the piperidine alkaloids was transferred to a round bottom flask after filtration, and chloroform was evaporated on a rotary evaporator at 57°C to produce the solid basic chloroform extract (BCE) of piperidine alkaloids from mesquite [32]. The BCE analysis by HPLC-MS identified juliprosopine (C₄₀H₇₆N₃O₂ [M+H]⁺, MM = 630.54) as major constituent and juliprosinine (C₄₀H₇₂N₃O₂ [M+H]⁺, MM = 626.49) as minor constituent. The BCE was weighed and added to the concentrate feed to obtain the 9.2, 18.4, and 27.6 mg kg⁻¹ diet DM. The levels of enriched mesquite piperidine alkaloid extract (MPA) used were based on *in vivo* experiments with lambs where concentrations in the diets ranged from 2.3 to 31.5 mg kg⁻¹ DM.

2.3 Sample processing and laboratory analyses

Chemical analyses of the ingredients, experimental diets, residual feed, and feces were carried out according to the analytical procedures of the Association of Analytical Communities [33] following the grinding of the samples in Wiley knife mills (A.H. Thomas, Philadelphia, PA) with a 1-mm sieve. The dry matter (DM, method 967.03), crude protein (CP, method 981.10), ether extract (EE, method 920.29), ash (method 942.05), and total nitrogen contents were determined.

The levels of neutral detergent fiber (NDF), samples were treated with thermostable alpha-amylase, without the use of sodium sulfite and corrected for residual ash according to the methodology proposed by [34]. For the determination of acid detergent lignin (ADL), the ADF residue was treated with 72% sulfuric acid based on the methodology described by [35] in which ADF residue was obtained by sequential analysis. The hemicellulose content was determined as the difference between NDF and ADF, and the cellulose content was determined as the difference between ADF and ADL. The levels of nonfibrous carbohydrates were calculated according to the methodology proposed by [36].

The total digestible nutrients (TDN), digestible (DE), and metabolizable energy (ME) contents to formulate the diet was estimated according to equations described by [37]. The TDN obtained in the digestibility assay was calculated according to [38]. The TDN values were converted into DE and ME, using the equations suggested by [37].

2.4 Evaluation of intake, digestibility, and live weight gain

The daily individual intakes (DMI) were measured over 89 days of supply of the experimental diets, by subtracting the amount of feed offered and refused. The amount of feed was adjusted daily, with the acceptable refusal amount corresponding to 10% of the total amount supplied to ensure *ad libitum* intake. The animals were weighed at the start, every 29 days, and at the end of the experiment. At the beginning of the experimental period, the animals were subjected to a 16-h solid fast and weighed to determine initial body weight (IBW). Total weight gain (TWG) was estimated as the difference between final body weight (FBW) and initial body weight (IBW): $TWG = (FBW - IBW)$. Average daily gain (ADG) was calculated by dividing TWG by the total number of days in the experiment: $ADG = TWG/Days$ in feedlot. Finally, feed conversion ratio was calculated as the ratio between dry matter intake ($kg\ day^{-1}$) and TWG ($kg\ day^{-1}$).

Apparent total digestibility of nutrients was estimated by total collection of feces during 3 days for each range of 25 days, in growing goats with a fecal bag attached to them. The feces were sampled after homogenization. A composite sample of feces, based on dry weight, from collection days for each animal and sub-period was prepared for chemical analysis. During the total tract digestibility trial representative samples of supplied hay and concentrate, and residual feed was collected within 3 days and composed for each animal and sub-period for chemical analysis. The nutrient digestibility coefficients (DC) were calculated as proposed by [39], as the ratio between the ingested amount of a nutrient and its excretion in the feces: $DC (\%) = ((Nutrient\ intake, in\ g - nutrient\ excreted\ in\ the\ feces, in\ g) / Nutrient\ intake, in\ g) \times 100$.

2.5 Microbial protein synthesis and N balance

On the 25th day of each experimental sub-period, urine was collected as a urine spot sample by spontaneous urination from each animal, approximately 4 h after the morning meal [40]. These samples were intended for the quantification of the urinary concentrations of urea, creatinine, and uric acid, using commercial kits of Bioclin[®] (Delft, the Netherlands). Xanthine-hypoxanthine and allantoin were determined according to procedures described by [41]. The urine volume of each animal was estimated with daily creatinine excretion (DCE) divided by creatinine concentration determined in urine spot samples. DCE in the different experimental diets was obtained in another assay with five animals of the same genetic group, in metabolic cages, distributed in a 5×5 Latin square, and feeding the same experimental diets. The values used were 16.03, 14.05, 17.59, 18.4, 21.26, and 14.87 $mg\ kg^{-1}\ BW$, respectively for MON, 0, 9.2, 18.4, 27.6 $mg\ kg^{-1}$ MPA diets.

The amount of absorbed microbial purines ($mmol\ day^{-1}$) and the intestinal flow of microbial nitrogen ($g\ MN\ day^{-1}$) were estimated from the excretion of total purines ($mmol\ day^{-1}$), using equations proposed by [42], for growing goats. The microbial efficiency was obtained by dividing the microbial protein synthesis ($g\ day^{-1}$) by the intake of TDN ($kg\ day^{-1}$) [43-44, 37].

2.6 Statistical analysis

Data analysis was performed using the GLM procedure of SAS statistical software version 9.1 (SAS Institute, Inc. Cary, NC). The average data from the three sampling sub-periods were used for apparent digestibility, microbial protein synthesis, and nitrogen balance, and the normality of variance was verified. The intake data for each animal were obtained by dividing the total ingested nutrients amount by the days of the experiment duration (89 days).

The contrast was applied to compare the means observed between the diets with MON *versus* MPA concentrations. The polynomial contrasts were performed for the linear (L) and quadratic (Q) components. For the dependent variables whose polynomial contrasts were significant, regression analysis of linear (L) or quadratic (Q) effects was performed according to

the MPA concentrations in diets (0; 9.2, 18.4, and 27.6 mg kg⁻¹ DM). The critical level of significance adopted was $P < 0.05$ and for tendency was $0.05 < P < 0.10$.

III. RESULTS

3.1 Evaluation of intake, digestibility, and live weight gain

The intakes of dry matter (DM), organic matter (OM), and neutral detergent fiber corrected for ash and protein (NDF_{ap}) were not affected ($P > 0.05$) by levels of enriched mesquite piperidine alkaloid extract (MPA) in diets. There was a tendency ($P = 0.051$) to reduce the NDF_{ap} (g kg⁻¹ BW) in diets with MPA compared to the diet with monensin (MON) (Table 2).

The levels of MPA increased 0.086 g kg⁻¹ BW^{0.75} the crude protein (CP, $P = 0.0004$) intake for each unit of MPA inclusion, which showed lower ($P = 0.004$) mean (10.64 g kg⁻¹ BW from 9.2 to 27.6 mg kg⁻¹ MPA) than MON diet (11.57 g kg⁻¹ BW). The intake of non-fiber carbohydrates (NFC, $P = 0.035$) increased 0.083 g per kg BW^{0.75} for each unit of MPA inclusion in the diets, and the mean (9.2 to 27.6 mg kg⁻¹) MPA was similar ($P > 0.10$) to the MON diet.

There was a quadratic response for the intakes (g day⁻¹) of ether extract (EE, $P = 0.023$), total digestible nutrients (TDN, $P = 0.008$), digestible (DE, $P = 0.008$) and metabolizable energy (ME, $P = 0.007$), with a maximum at 10.5, 15.3, 17.3, and 17.4 mg kg⁻¹ MPA, respectively. These variables did not differ from MON (Table 2).

There was no difference ($P > 0.10$) among diets with MPA when compared to MON for the digestibility of DM, OM, CP, and NDF_{ap}. For EE, MPA diets provided the highest mean compared to MON diet ($P = 0.001$) and quadratic effect ($P = 0.034$) with the MPA levels.

The CP and NFC digestibilities increased with the MPA levels showing an increment of 0.22 g 100 g⁻¹ ($P = 0.003$) and 0.14 g 100 g⁻¹ ($P = 0.040$) for each unit of MPA inclusion in the diet, respectively (Table 3). The average CP digestibility did not differ ($P > 0.10$) between diets with MPA and MON and the NFC digestibility tended to be higher for diets with MPA ($P = 0.093$).

Diets supplemented with MPA changed the daily weight gain (DWG) in growing goats ($P = 0.044$) with the maximum point at 14.8 mg kg⁻¹ MPA, showing a 0.2 g reduction for each unit of MPA inclusion after the peak. The average DWG of growth goats fed diets supplemented with MPA did not differ from the diet with MON, and both additives promoted a 28% increase in DWG compared to diet without additives (Table 3). The feeding efficiency (FEF) tended ($P = 0.067$) to be maximum with 15.6 mg kg⁻¹ MPA in the diet.

3.2 Microbial protein synthesis and N balance

There were no differences ($P > 0.10$) among diets containing MPA compared to the diet with MON for the synthesis of microbial nitrogen (Table 4). But, the efficiency of microbial protein synthesis (MEF) showed a decreasing linear effect ($P = 0.043$), in which the mean for diets with MPA was equal to 87.35 g kg⁻¹ of TDN. However, there was no difference ($P > 0.10$) between MPA and MON.

There was a linear increase of the nitrogen intake (NI, $P = 0.022$) and of the amount of digested N (DN, $P = 0.017$) with the MPA levels and both variables did not differ from the MON diet. The excretion of N in the feces (FN) varied quadratically ($P = 0.025$) with a minimum point at 13.3 mg kg⁻¹ MPA. The urinary N (UN) was no changed ($P > 0.10$) according to the MPA levels and it was greater ($P = 0.038$) for MPA diets than the diet with MON (Table 4). The plasma urea N (PUN) concentration peaked ($P = 0.019$) at 16.9 mg kg⁻¹ MPA. There was an increase ($P = 0.006$) in the amount of retained N (RN) with MPA levels and the diets with MPA were lower ($P = 0.040$) than the MON diet.

The proportion between the amounts of RN and N intake (g RN 100 g⁻¹ NI) tended ($P = 0.057$) to be lower for MPA diets in comparison with MON diet. The RN to digested N ratio (g RN 100 g⁻¹ DN) was lower ($P = 0.020$) for MPA diets compared to MON diet (Table 4). However, there was an improvement ($P = 0.043$) in the conversion of RN in DWG (g 100 g⁻¹) with the MPA in comparison with MON diet.

TABLE 2
INTAKE AND DIGESTIBILITY OF NUTRIENTS IN GROWING GOATS FED DIETS WITH CONCENTRATIONS OF ENRICHED MESQUITE PIPERIDINE ALKALOID EXTRACT (MPA) OR MONENSIN (MON)

Item	Experimental diet (mg kg ⁻¹ DM)					SEM	P-Value		
	MON	MPA					MON vs MPA	L	Q
	2.7	0	9.2	18.4	27.6				
DM (kg day ⁻¹)	0.91	0.77	0.86	0.90	0.86	0.02	0.497	0.087	0.053
OM	0.88	0.74	0.77	0.84	0.75	0.03	0.252	0.772	0.402
CP	0.15	0.11	0.12	0.15	0.14	0.004	0.260	0.002 ¹	0.095
EE	0.03	0.03	0.03	0.03	0.02	0.001	0.977	0.185	0.023 ²
NDF _{ap}	0.28	0.23	0.22	0.26	0.22	0.006	0.011	0.847	0.222
NFC	0.38	0.31	0.37	0.38	0.39	0.009	0.977	0.005 ³	0.201
TDN	0.64	0.56	0.65	0.66	0.63	0.01	0.909	0.037	0.008 ⁴
DE (MJ day ⁻¹)	11.88	10.13	12.05	12.18	11.67	0.06	0.909	0.037	0.008 ⁵
ME	10.29	8.79	10.54	10.63	10.17	0.05	0.801	0.033	0.007 ⁶
DM (g kg ⁻¹ BW)	30.22	27.76	29.36	30.32	29.26	0.69	0.719	0.231	0.161
NDF _{ap}	9.31	8.23	7.41	8.98	7.80	0.23	0.051	0.857	0.635
CP (g kg ⁻¹ BW ^{0.75})	11.57	8.87	9.55	11.53	10.84	0.28	0.004	0.0004 ⁷	0.151
EE	2.05	2.25	2.43	2.28	1.88	0.80	0.448	0.116	0.081
NFC	29.35	26.08	29.84	29.38	30.54	0.71	0.743	0.035 ⁸	0.324
TDN	49.69	45.43	51.31	51.98	50.11	0.10	0.547	0.069	0.018 ⁹

¹Y= 0.11 + 0.0013 X; ²Y= 0.027 + 0.00063 X - 0.00003 X²; ³Y= 0.32 + 0.0028 X; ⁴Y= 0.56 + 0.0122 X - 0.0004 X²; ⁵Y= 10.19 + 0.2497 X - 0.0072 X²; ⁶Y= 8.85 + 0.2261 X - 0.0065 X²; ⁷Y= 9.01 + 0.0858 X; ⁸Y= 29.09 + 0.0834 X; ⁹Y= 45.56 + 0.7917 X - 0.0229 X²

NDF_{ap}, Neutral detergent fiber correct for ash and protein; NFC, Non-fiber carbohydrates; TDN, Total digestible nutrients; DE, digestible energy; ME, metabolizable energy; SEM, standard error of the mean; L, linear; Q, quadratic.

TABLE 3
DIGESTIBILITY OF NUTRIENTS AND PERFORMANCE OF GROWING GOATS FED DIETS WITH CONCENTRATIONS OF ENRICHED MESQUITE PIPERIDINE ALKALOID EXTRACT (MPA) OR MONENSIN (MON).

Item	Experimental diet (mg kg ⁻¹ DM)					SEM	P-Value		
	MON	MPA					MON vs MPA	L	Q
	2.7	0	9.2	18.4	27.6				
Apparent digestibility (g 100 g ⁻¹ DM)									
DM	85.11	83.13	82.84	80.65	85.59	0.85	0.275	0.414	0.136
OM	85.15	81.02	88.02	83.19	83.57	1.14	0.815	0.807	0.204
CP	82.77	77.75	79.60	79.49	84.08	0.80	0.341	0.003 ¹	0.429
EE	65.45	68.54	72.24	74.75	71.75	0.82	0.001	0.067	0.034 ²
NDFap	66.65	64.34	68.12	69.14	69.06	1.20	0.567	0.196	0.406
NFC	83.85	83.62	85.05	88.37	86.81	0.66	0.093	0.040 ³	0.285
TDN	69.74	72.93	76.79	74.78	72.45	9.16	0.065	0.691	0.158
Growth performance (kg)									
IBW	22.56	21.98	21.92	21.78	21.92	-	-	-	-
FBW	37.74	33.07	37.36	37.26	36.03	0.93	0.742	0.354	0.196
TWG	15.18	11.09	15.44	15.47	14.11	0.71	0.926	0.190	0.070 ⁴
DWG	0.171	0.125	0.173	0.174	0.159	0.01	0.944	0.140	0.044 ⁵
FEF (g 100 g ⁻¹)	21.44	19.36	24.16	23.88	21.72	0.84	0.429	0.412	0.067 ⁶
¹ Y = 77.52 + 0.2199 X; ² Y = 68.41 + 0.6397 X - 0.0186 X ² ; ³ Y = 84.02 + 0.140 X; ⁴ Y = 11.24 + 0.5643 X - 0.0169 X ² ; ⁵ Y = 0.127 + 0.0059 X - 0.0002 X ² ; ⁶ Y = 19.52 + 0.6413 X - 0.0206 X ²									
<i>IBW, Initial body weight; FBW, Final body weight; TWG, Total weight gain; DWG, Daily weight gain; FEF, feeding efficiency: daily weight gain ÷ digestible dry matter intake; SEM: standard error of the mean; L, linear, Q, quadratic.</i>									

TABLE 4
NITROGEN BALANCE AND MICROBIAL SYNTHESIS IN GROWING GOATS FED DIETS WITH CONCENTRATIONS OF ENRICHED MESQUITE PIPERIDINE ALKALOID EXTRACT (MPA) OR MONENSIN (MON).

Item	Experimental diet (mg kg ⁻¹ DM)					SEM	P-value		
	MON	MPA					MON vs MPA	L	Q
	2.7	0	9.2	18.4	27.6				
Microbial nitrogen (g day ⁻¹)	8.01	9.35	8.92	10.47	7.79	0.35	0.296	0.333	0.123
MEF (g CP kg ⁻¹ TDN)	80.85	111.2	84.75	98.87	78.42	3.83	0.702	0.043 ¹	0.534
Nitrogen intake (NI, g day ⁻¹)	24.37	17.35	19.72	23.75	22.11	0.86	0.240	0.022 ²	0.245
Nitrogen excretion (g day ⁻¹)									
Feces	4.30	4.10	5.16	5.70	3.79	0.29	0.454	0.883	0.025 ³
Urine	1.22	3.23	2.00	2.85	2.16	0.29	0.038	0.751	0.865
Plasma urea nitrogen (mg dl ⁻¹)	14.79	9.43	13.12	14.18	11.97	0.65	0.416	0.150	0.019 ⁴
Digested nitrogen (DN)									
g day ⁻¹	20.07	13.25	14.56	18.05	18.32	0.83	0.138	0.017 ⁵	0.752
g 100 g ⁻¹ NI	81.44	75.03	63.36	74.88	82.46	1.85	0.069	0.004 ⁶	0.725
Retained nitrogen (RN)									
g day ⁻¹	18.85	10.02	12.56	15.20	16.17	0.87	0.040	0.006 ⁷	0.619
g 100 g ⁻¹ NI	75.56	62.50	72.65	63.67	72.96	1.78	0.057	0.136	0.898
g 100 g ⁻¹ DN	93.86	86.10	84.49	87.02	88.35	1.20	0.020	0.427	0.564
RNC (g 100 g ⁻¹)	11.83	9.21	7.21	8.83	10.13	0.57	0.043	0.168	0.131
¹ Y= 105.94 - 0.9154 X; ² Y= 17.99 + 0.199 X; ³ Y= 4.06 + 0.236 X - 0.0089 X ² ; ⁴ Y= 14.02 + 0.199 X; ⁵ Y= 13.24 + 0.2033 X; ⁶ Y= 68.86 + 0.368 X; ⁷ Y= 10.32 + 0.229 X MEF, Microbial efficiency; CP, microbial crude protein; TDN, total digestible nutrients intake; RNC, retained nitrogen conversion: retained nitrogen ÷ daily weight gain; SEM, standard error of the mean; L, linear; Q, quadratic									

IV. DISCUSSION

The supplementation of the diets with MON or MPA did not affect the DM intake. However, both additives increased the intakes of CP, NFC, TDN, DE, ME and the supplementation with MON increased the NDF_{ap} intake, indicating selective behavior. The MPA diet increased CP, EE, and NFC digestibilities resulting in greater TDN intake than the diet without additives.

The maintenance of the digestibility of DM and OM in diets with MON or MPA levels may have been the factor that contributed to the similarities in DM and OM intakes of diets with additives. On the other hand, when considering the diet without additives, the similar response for DM intake did not correspond to the composition of the ingested diets with MON or MPA due to feed selection. The use of a high concentrate proportion may cause digestive disorders, resulting in decreased intake, a fact that did not occur since the DM intake was reached due to the adaptation of the animals to the roughage and concentrate ratio (20: 80).

The polynomial contrast showed a significant quadratic component of the effects of MPA levels on the EE digestibility, consistent with the effect on its intake. Likewise, the linear increase in the digestibility of CP and NFC with the use of MPA levels reflects the increased intake of these nutritional components. The intake composition expresses as TDN and protein ratio (g kg^{-1} of diet DM) was: MON = 4.2; without additive = 5.1; MPA 9.2 mg kg^{-1} = 5.5; 18.4 mg kg^{-1} = 4.5, and 27.6 mg kg^{-1} = 4.4. The higher intake of CP and NFC indicates that there was a change in the roughage and concentrate ratio of the diet consumed, which justifies the greater intake and digestibility of these components. The variation in the composition of the consumed diet is probably due to the ability of goats to select during prehension [45]. [28] observed that MPA at 9.2 mg kg^{-1} increased the utilization of NFC in lambs fed diets with MPA levels (2.3; 4.6 and 9.2 mg kg^{-1} of diet DM) composed of 60 % concentrate and 40 % roughage.

The fact that MON and MPA did not affect the NDF_{ap} digestibility, associated with the stable DM and OM intake and digestibility indicates that the amounts used of the additives maintained the conditions for fiber degradation in the rumen. This is an important aspect of evaluating an additive, especially using low-quality roughage. Thus, it indicates responses quite similar to monensin, which are maintenance of rumen pH more favorable to cellulolytic bacteria and amylolytic bacteria that provide degradation products used by gram-negative cellulolytic bacteria [2, 46-47].

In ruminants fed high concentrate diet, the fiber digestibility has often been increased by the inclusion of additives, and this increase may be the result of changes in the microbiome activity and/or the longer retention of fiber in the rumen, which favors the extension of microbial digestion [2]. Conversion of rumen digested energy to microbial protein is less efficient with reduced rumen turnover, but the increased extent of rumen digestion may partially compensate to maintain microbial protein output from the rumen [48].

The estimated CP intake for growing goats with average body weight (BW) of 25 kg is approximately 0.115 kg day^{-1} , with an estimated daily gain of 180 g [30]. In diets with MON and MPA at 18.4 mg kg^{-1} , there was an average daily CP intake of 0.150 kg, and in growing goats showed similar daily weight gain (DWG). For the diet containing MPA at 9.2 mg day^{-1} , the daily intake of CP was 0.120 kg and the goats also reached similar DWG. This response showed that the digestion and/or utilization of diet protein depend of the MPA dose and the minimum concentration effective of MPA for maximum DWG was 9.2 mg kg^{-1} .

In contrast, the diet without additives was no efficient, causing a CP intake of 0.110 kg day^{-1} with DWG of 0.125 kg, although the ME intake was closer to the recommended by [30]. The ME requirement recommended is 7.82 MJ day^{-1} by the [30] for crossbred kids goats with 25 kg BW and an estimated daily gain of 180 g.

Commonwealth Scientific and Industrial Research Organisation (CSIRO) defined the potential for food intake as the amount of food eaten when offered *ad libitum*, and the animal can select a diet with a minimum DM digestibility of 80 % or with a minimum concentration of 11 MJ kg^{-1} of DM [49]. In this study, it was observed the average value of DM digestibility above 80%, and the ME content, which averaged 11.25 MJ kg^{-1} for diets with MPA. Consistently, there was a linear increase of the CP and NFC intakes with MPA levels indicating diet selection, probably, as a consequence of a change in the rumen digestion [50].

The feeding efficiency (FEF) tended to achieve the peak at approximately 15 mg kg^{-1} MPA and the higher daily weight gain (DWG) at this point confirms the potential use of MPA as an additive for growing goats fed the diet with a high concentrate proportion.

In this study, the response of DWG was due to the use of additives, mainly as a reflection of the improvement in the intake of digestible and metabolizable energy from diets. It was demonstrated that the addition of MPA was effective to increase DWG in goats consuming 20% Tifton 85 hay. There was a plateau for DWG between 9.2 and 18.4 mg kg⁻¹ MPA levels, demonstrating that this range of MPA inclusion in diets promotes the best response in growing goats.

The 27.6 mg kg⁻¹ MPA did not promote an increase in DWG, probably in response to factors affecting the digestive processes that changed the energy and crude protein intakes. [13] reported that juliprosinine and juliprosopine interact with catalytic site residues of enzymes in pH 8.0, however, the rumen and gut have a lower pH than this resulting in less stable interactions. The lower pH could favor the action of piperidine alkaloids of molecular mass above 600 Da to act as an ionophore in the membrane of gram-positive bacteria and not of gram-negative bacteria which present the outermost membrane barring the access of ionophores to the plasmatic membrane [11, 51].

The MPA ranging from 9.2 to 18.4 mg kg⁻¹ tended to increase the feeding efficiency (FEF). [28] reported that MPA (2.3 to 9.2 mg kg⁻¹) improved the feed conversion compared to MON in lambs fed diets composed of 40% Tifton 85 hay and 60% concentrate. Hence, the authors suggested more studies with higher concentrations would be promising.

In high concentrate diets, MON generally maintains or increases the weight gain and improves feed conversion [52]. In this study, even though there was a tendency to increase the FEF in growth goats fed diets with MPA levels, it was found that the increased DWG with the same DM intake as a consequence of the improvement in the energy and protein utilization of the diets.

For MON and MPA at 27.6 mg kg⁻¹, there was a higher amount of dietary retained nitrogen (RNC) for every 100 g of weight gain. Possibly, the largest fraction of the amount of retained N was used in other metabolic routes that use amino acids than the protein synthesis to be deposited in the gain of BW due to the ME intake does not to follow the increase of CP intake. Among the major factors affecting the post-rumen N efficiency is the amount of energy available [53]. Increasing the supply of post-rumen energy substrates while maintaining protein supply improves the efficiency of utilization of amino acid for gain; and thus reduces the output of N in urine [54].

There was a decrease in the concentration of plasma urea nitrogen (PUN) and the feces N excretion reduced in the highest level of MPA, without observing a change in the excretion of N in the urine. Thus, the increase in the amount of digested N and retained N showed that probably there was a reduction in the degradation of dietary protein in the rumen, and greater use of dietary amino acids in the intestine with 27.6 mg kg⁻¹ MPA [55]. [56] reported that monensin decreased bacterial N and more dietary protein reached the abomasum of steers adapted to monensin. Other reports support a higher ruminal escape effect of dietary protein by monensin [57; 2].

The MPA at 27.6 mg kg⁻¹ showed an effect of less protein degradation in the rumen, due to the lower PUN value, and the diets with MPA at 9.2 and 18.4 mg kg⁻¹ were more efficient in the synthesis of microbial protein, consistent with that observed by [28] using the maximum level of 9.2 mg kg⁻¹ MPA for sheep. The increase in microbial efficiency allows an increase in the availability of microbial protein to be absorbed in the intestine, supplying, thus, the requirements of growing animals [58]. The higher microbial efficiency of diets with MPA at 9.2 and 18.4 mg kg⁻¹ compared to 27.6 mg kg⁻¹ may have contributed to the higher DWG in growing goats fed with the first two diets.

The MON diet provided a rise of 33% in retained nitrogen (RN) when compared with the 9.2 mg kg⁻¹ MPA diet, however, both showed similar DWG. [28] observed that the N intake was not changed in lambs, as it was consistent with similar CP levels in the diets. At that study, for the diet with MPA in the highest dose of 9.2 mg kg⁻¹, the effect of greater feces N excretion caused a reduction in the proportion of retained N relative to ingested N. This finding was supported in the present study, which was noted a tendency of decrease in the RN proportion (g 100 g⁻¹ NI) in MPA diets compared to MON diet.

The RN proportional to the ingested N can predict the efficiency of the use of N in the animal organism [59]. It is more strongly associated with the supply of N than with the energy content of the diet, and it is amplified by the improvement in the conditions of protein status in the animal organism [60]. There was a linear increase in the N intake with the MPA levels, consequently, the DN and RN proportional to the ingested N also increased. In addition, these variables did not differ from MON diet. The DN proportional to the N intake was similar between the MON diet and 27.6 mg kg⁻¹ MPA diet, indicating that both take action equally in the protein digestion.

The protein status refers to the qualitative and quantitative availability of nitrogen compounds for all physiological functions in tissue metabolism, including functions associated with energy metabolism [60]. However, the RN proportional to the

digested N and the RN conversion (RNC) was lower with MPA at 9.2 mg kg^{-1} than with MON in the diet, possibly, due to the dose-response effect to the MPA. However, both additives were comparables to nitrogen excretion (feces and urine) and DWG. The 18.4 mg kg^{-1} MPA increased the N intake and in comparison with MON, it showed higher urinary N excretion, a tendency to reduce RN proportional to the ingested N, and similarity in DWG. In contrast, the 27.6 mg kg^{-1} MPA increased the N intake, digested nitrogen, retained nitrogen and it reduced the microbial efficiency and also DWG.

Probably, in the rumen environment, the additives performed as modulators of the fermentation with metabolic routes that produce less methane and deamination of amino acids resulting in lower energy loss or spilling energy [1]. The effectiveness of an additive is measured using productive and metabolic parameters. The monensin reduces protein degradation with the decrease of rumen N-NH_3 concentrations and increases the intestinal digestion of dietary protein [2, 61]. As long as it does not compromise the efficiency of microbial synthesis, it can be beneficial, since the excess of rumen N-NH_3 has a high energy cost in the transformation of urea in the liver [62], reducing the diet energy yield [63]. Also, the reduction in rumen concentration of N-NH_3 can occur due to the availability of energy in the rumen, which allows greater use of ammonium for microbial growth, with a consequent reduction in urine urea loss [59].

The excretion rates of nitrogen compounds in the urine and feces of ruminants are associated with the amount of DN [64]. [54] related that the variation in dietary N intake will particularly affect the excretion of urinary N, which is much more vulnerable to losses than is fecal N. The quantity of N excreted in urine varies widely. Urinary N excretion, in particular, that of urea N, is decreased upon reduction of dietary N intake or an increase in the supply of energy to the rumen microorganisms and to the host animal itself.

The 27.6 mg kg^{-1} MPA showed a reduction in the concentration of PUN possibly due to the action to limit available ammonia for bacterial growth as a consequence of increased escape of dietary protein from the rumen. Hence, the greater N intake has resulted in a value like MON for retained nitrogen conversion per 100 g weight gain (RNC). Possibly, the growth goats fed the diet with MPA at 27.6 mg kg^{-1} showed lower DWG response than MON and 9.2 to 18.7 mg kg^{-1} MPA as a result of a decrease in DE and ME intake. On the other hand, the lower dose of MPA showed better efficiency of energy and protein utilization due to greater microbial efficiency and daily weight gain with lower CP intake. Consistently, [29] proposed a CP reduction, from 16% to 13%, in the diet for lambs fed diet constituted of 33.3% Buffelgrass hay and 66.7% concentrate with 31.5 mg kg^{-1} MPA due to the rise of microbial efficiency in the rumen.

V. CONCLUSIONS

The doses of MPA between 9.2 to 18.4 kg^{-1} of feed dry matter for growing goats improve metabolizable energy intake and daily weight gain with the lower conversion of retained nitrogen than MON. To achieve a better description of the action mechanism of main alkaloids obtained from the alkaloid-rich fraction (juliprosopine and juliprosinine), additional studies on enzyme activity in the rumen and small intestine and microbial population are required.

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Impacts of Illegal Mining on Human Being, the Case of Huye District

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Abstract— *The current study was about the impacts of illegal mining on wellbeing of people. More specifically the study intended to:*

- (i) *identify illegal mining practices in Huye District;*
- (ii) *Assess the social, economic and environmental impacts of illegal mining on wellbeing of people of Huye District.*
- (iii) *Find out solutions to social, economic and environmental impacts of illegal mining on environment and wellbeing of people of Huye District.*

Keywords— *Huye District, Illegal Mining impact on Human.*

I. INTRODUCTION

Mining has been a most concern activity for economy's growth Worldwide. UNEP (2009) says that mining is an activity that involves digging of surface and subsurface for the purpose of exploiting and processing the minerals for just various uses. Gibson (2018) reveals that extracted minerals can be copper and nickel; precious metals like gold and silver; iron; uranium; coal; etc.

Mining can be legal or illegal depending on the status of rights owned by the miners. More specifically, Gibson et al, (2018) said that illegal mining refers to violations of both specialized laws and regulations for mining that protect the environment and human welfare. UNEP (2009) added that illegal mining for the most world countries is characterized by the lack of land rights, mining licenses and transportation permit.

Mineral resources belong to the government; and therefore they must be exploited by the miners with the legal rights regarding to the laws and regulations (Phillipe, 2019). Many specialists and policymakers confirm that illegal mining are seriously connected to the poverty for people living nearby the mineral areas; and this is mainly due to soil erosion, destruction of farms and crops thereby small agricultural harvest.

In Peru, researches indicate that illegal mining lead to heavy metal and chemical poisoning of environments (Fashola et al, 2016), and the erosion of soils (Dissanayake and Rupasinghe, 2017); hence, USAID in the partnership with Peru works towards this, through the rehabilitation of the affected land, preventing the expansion of illegal mining as well, into protected areas and other forested lands.

In USA illegal mining has been considered as a destructive agent. In light of this; Amazonian communities, their forests and ecosystems have been destroyed by illegal miners; and this lead to the signature of a Memorandum of understanding in 2017 between USA and Peru to combat illegal gold mining (Jene, 2019); this ensures long life of environmental ecosystems.

In India, illegal mining takes place in abandoned mining sites and its effects appear all over the whole region where human being encounters serious problems (Mari Hayman, 2013). UN (2018) reported that illegal mining leads to the blood diamond; that's conflict diamonds between rebels and international recognized governments. Mari Hayman (2013) says that blood diamonds are used to fund military actions in opposition to those governments. World diamonds council (2016) argued that the sale of blood diamonds support drug trafficking and terrorism which are deadly in nature.

In South Africa, illegal mining is a criminal and highly dangerous activity. The most concerned minerals are diamonds, chrome and coal (Mineral Council report, 2017). UN (2018) claims that illegal mining in South Africa deserves a whole range of negative social impact across the board; these are like death of illegal miners, volunteers in rescuing those who have been trapped underground. And this presents major threats to African governments in safeguarding their population (Mining Review Africa, 2019).

In DRC, much mining has been done illegally with a high level of child labor and workplace injury (Hanah, 2019). Sylvain (2018) said that some armed groups in DRC finance their operations through the illegal mining, thereby destructive conflicts in the region.

In Rwanda, illegal mining is carried out to extract cassiterite, Wolfram and Coltan. Rwanda Mines Petroleum and Gas Board (2018) asserted that people died, others get injured due the illegal mining activities. John Kanyangira (2019) suggested that security in mining sites must be granted to rescue the mining operators. RIB (2019) says that illegal mining is risky to people's lives.

In Southern Province areas where illegal mining is practiced include in Huye District; and minerals extracted are like coltan and Cassiterite. In Huye District, illegal mining is conducted in Rwaniro Sector where the minerals like coltan and cassiterite are mostly extracted (Viviane Irabizi, 2019).

II. MATERIALS AND METHODS

2.1 Research design

The design of a research is the combinations of methods you have chosen for empirical part of your study (Kenneth, 1978). Grinnell et al (1990) define research design as the comprehensive process of the study and the problem formulation through dissemination of findings. Under this research, descriptive research design was used to assess the impacts of illegal mining on the human being in environment in Huye District as the case study.

2.2 Descriptive research design

Descriptive research design was used in the current study. This was to provide essential information in need. It is however made by the qualitative and quantitative research approaches.

2.3 Qualitative research approach

Qualitative research approach seeks to provide an understanding of human experience, perceptions, motivations, intentions, and behaviors based on description and observation. Qualitative research is descriptive approach in which the researcher is interested in process, meaning, and understanding gained through words or pictures (Ramanath, 2010). The process of qualitative research is inductive in which the researcher builds abstractions, concepts, hypotheses and theories from the information gathered.

2.4 Quantitative research approach

Quantitative research approach emphasizes objective measurements and the statistical, mathematical, or numerical analysis of data collected through polls, questionnaires, and surveys, or by manipulating pre-existing statistical data using computational techniques. Quantitative research focuses on gathering numerical data and generalizing it across groups of people or to explain a particular phenomenon (Ramanath, 2010). This study will adopt Quantitative research approach.

2.5 Sampling design

2.5.1 Study population

A population is the mass of units of analysis about which, the researcher was measure his or her variables (Kenneth, 1978). According to Panneerselvam (2005), a study population refers to the total group of people from whom the information is needed. The study population comprises 230 persons of people around the mines in Kamwambi and Nyaruhombo cells, Rwaniro sector, Huye District.

2.5.2 Sampling techniques

Basically, sampling techniques are divided into probability and non-probability sampling. Probability sampling provides an equal opportunity for each and every element of the population being selected. This method utilizes some form of random

selection. But non- probability sampling does not involve random selection (Ramanath, 2010). In this context, purposive sampling and convenience sampling techniques was used to determine the representative sample size.

2.5.3 Purposive sampling

Purposive sampling is a sampling technique in which researcher relies on her own judgment when choosing members of population to participate in the study. Purposive sampling is a non-probability sampling method and it occurs when “elements selected for the sample are chosen by the judgment of the researcher. Researchers often believe that they can obtain a representative sample by using a sound judgment, which was result in saving time and money (Ramanath, 2010). The researcher used purposive sampling technique since she has deliberately decided to consider only people respondents with the needed information about the topic being investigated.

2.5.4 Convenience sampling

The population or universe represents the entire group of units which is the focus of the study. Thus, the population could consist of all the persons in the country, or those in a particular geographical location, or a special ethnic or economic group, depending on the purpose and coverage of the study (Black, 2010).

The researcher was used convenience sampling technique while collecting information related to the impacts of illegal mining on human being in environment. The technique has been chosen because the researcher was collected the information from only available respondents depending on the time and space.

2.6 Sample size

A sample is a portion of the population selected to achieve the objectives of the research. Then, the sample size of the current study was calculated using the Slovin’s formula.

If you take a population sample, you must use a formula to figure out what sample size you need to take. Slovin’s formula is essential to figure out what sample size you need to take, which is written as

$$n = \frac{N}{(1 + Ne^2)}$$

Where n = Sample Size,

N = Total population,

e = Error tolerance,

Assume that a confidence level of 90 percent (which give a margin error of 0. 1 was used).

$$n = \frac{N}{(1 + Ne^2)} = \frac{230}{(1 + 230 * (0.1)^2)} = 69.69 = 70 \text{ Respondents}$$

The sample size was computed using the formula of Slovin and the study population made of population nearby the mining areas.

2.7 Study Area Description

This part focuses on presentation of Rwaniro Sector, Huye District as the study area, the precise area where mining activities present.

The current study has been conducted on illegal mining found in Rwaniro Sector, Huye District and Southern Province, Rwanda.

Huye is one of eight Districts that make up Rwanda’s Southern Province. It has a total surface area of 581.5 square Kilometers. It has fourteen sectors and 77 Cells with a total of 509 Umudugudus in total. The district has a population of 314,022 inhabitants with an average of 540 inhabitants per square kilometer. Like some other Districts, Huye District has minerals being extracted for economic purpose. However, the main minerals exploited in this District are Coltan and Cassiterite; and they are found in Rwaniro Sector (The New Times, 2017). Gatare Francis (2018) exhorts the government officials specifically in Huye Districts to join efforts in combating illegal mining which appears to the sector; and cause disastrous deaths due to illegal mining practices.

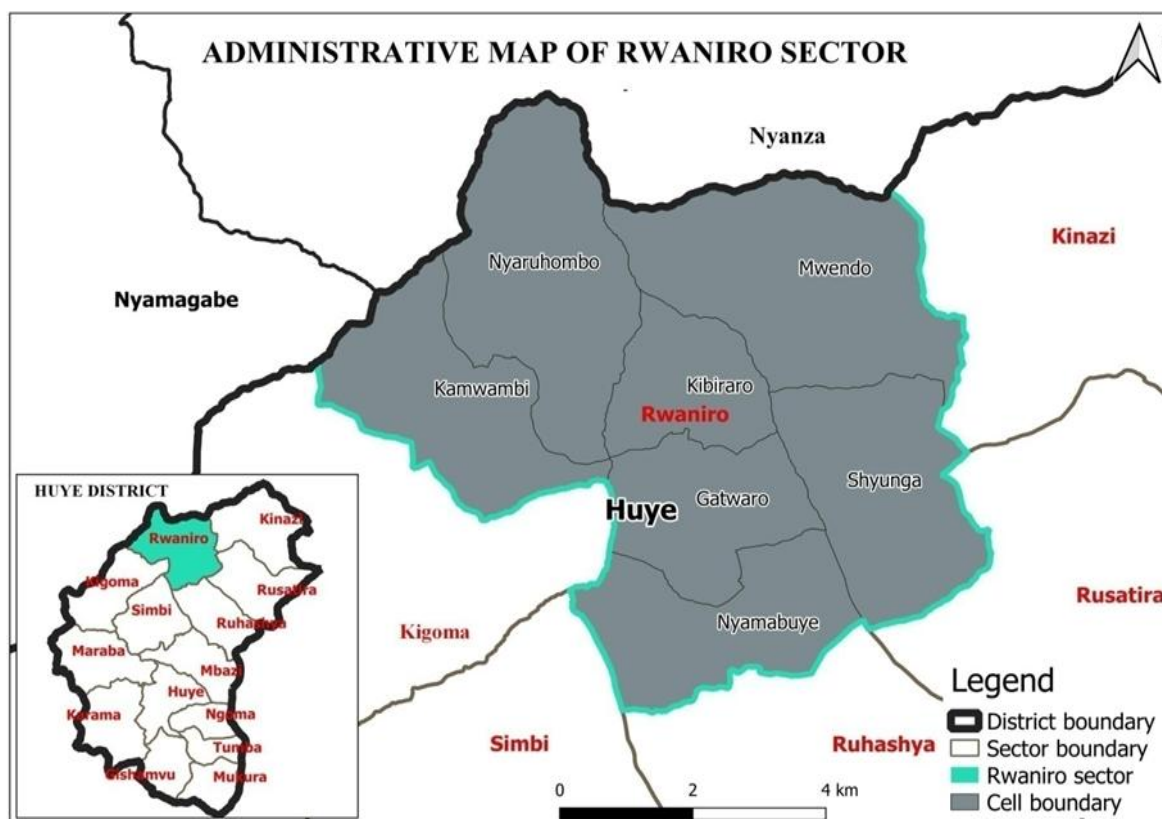


FIGURE 1: Map of Rwaniro sector, Huye District

Source: Author by using ArcGIS

2.8 Data Collection Procedures

2.8.1 Types of data

During this study, the researcher collected primary data through self-designed questionnaire.

2.8.2 Categories of data

For this study, the data collected were categorized in two categories. The researcher seeks to collect ordinal data. Ordinal data were collected through a questionnaire addressed to the population around the mines in the District under study.

For primary data collection, the researcher was used a structured questionnaire. Regarding secondary data, the researcher was used reports from Huye District.

2.8.3 Pre- testing

The pre-testing is the trial run of the questionnaires on a small sample of respondents to identify and eliminate potential problems (Maholtra, 2006). It has a role in ensuring the research instruments as a whole functions well in order to eliminate variation understanding and interpretation of the questionnaire in terms of ambiguity (Bryman and Bell, 2007). Before finalizing a questionnaire to be used to collect data, a pre-test has to be done to eliminate several mistakes that cannot be easily identified. Conducting a pre-testing study gives an advanced warning to the researcher about where the main research could fail, where research protocols may be followed and even whether the proposed methods or instrument are appropriate or complicated (Van & Hundley, 2001). For the purpose of this study, the questionnaire was pre-tested with 10 respondents. Thereafter changes were made about the wording, sequence and language of the questionnaire.

2.8.4 Pilot study

A pilot study is like a small version of the full study. In this phase, the questionnaire was piloted with 10 selected respondents from Muhanga District Mines which is not part of the study. Changes were made to the questionnaire in order to prepare the main survey instrument. The pilot study was initially undertaken to determine the reliability of the questionnaire. Statistical methods were used to calculate the reliability of multiple item measures.

2.9 Data Analysis Procedure

2.9.1 Data processing and analysis

2.9.1.1 Data processing

Raw data was transformed into meaningful interpreted report using different techniques. In order to get quality information; there were needed for standard checking so that the researcher could end up with realistic data, which clearly reflected the depicted situation.

Stand checking was done through editing; coding and tabulation. This was done in order to reduce detailed data to manageable proportions.

2.9.1.2 Editing

According to Sekaran (2003), “editing is the process whereby errors in completed interview schedules and questionnaires are properly analyzed, scrutinized and verified in order to avoid errors and repetitions”. This type of data processing made the analysis simple and easy to the researcher.

2.9.1.3 Coding

According to Kenneth (1978), coding refers to “assigning a symbol or a number to a response for identification purposes. The purpose of coding in research is to put forward by Panneerselvam (2005) who argued that it is to “classify the answers to questions into meaningful categories as so to bring out their essential pattern”

2.9.1.4 Tabulation

Tabulation is the process of summarizing raw data and displaying the same in the form of statistical tables for further analysis. In a broader sense, tabulation is an orderly arrangement of data in columns and rows (Kothari, 2004).

2.9.2 Statistical treatment of data

In this study, both Excel and SPSS were used in processing and analysis of data for the presentation of findings, analysis and interpretation. The presentation focused on the research questions. The kind of statistical treatment depends upon the nature of the problem, especially the specific and the nature of data gathered. For this purpose, Karl Pearson coefficient was used to analyze the relationship or correlation between the variables under study.

Description and formula: In order to determine how strong the relationship is between two variables, a formula must be followed to produce what is referred to as the coefficient value.

The coefficient value can range between -1.00 and 1.00 . If the coefficient value is in the negative range, it means that the two variables are negatively correlated, or as one value increases, the other decreases. If the value is in the positive range, it means that the two variables are positively correlated or both values increase or decrease together.

$$r = \frac{N \sum XY - (\sum X)(\sum Y)}{\sqrt{[N \sum X^2 - (\sum X)^2][N \sum Y^2 - (\sum Y)^2]}}$$

Where: X and Y are variables, N: Sample size.

TABLE 1
INTERPRETATION OF PEARSON CORRELATION COEFFICIENT

1	$r = +/-1$	Perfect Correlation
2	$+/- 0.9 \leq r < +/- 1$	Strong Correlation (Very High)
3	$+/- 0.7 \leq r < +/- 0.9$	High Correlation
4	$+/- 0.5 \leq r < +/- 0.7$	Moderate Correlation
5	$r < +/- 0.5$	Weak (Low) Correlation
6	$r = 0$	Absence of Correlation

Source: Anand, 2010

2.10 Data analysis

Data collected were analyzed using tables, percentages and statistic tools such as Pearson Correlation.

III. RESULTS AND DISCUSSION

3.1 Results

3.1.1 Introduction

This part covers the results of the study, it started by describing the profile of the respondents and the findings of the research with respect to the following specific objectives: To identify illegal mining practices in Huye District ,to assess the social, economic and environmental impacts of illegal mining on wellbeing of people of Huye District, Rwanda and finding out solutions to social, economic and environmental impacts of illegal mining on environment and wellbeing of people of Huye District, Rwanda.

3.1.2 Respondents Identification

The survey included information on sex, age group, approximation of years of living near mining areas, approximation of years of living near main road used by trucks transporting extracted minerals, frequency of visiting the soil mining area and activities normally done in the soil mining area.

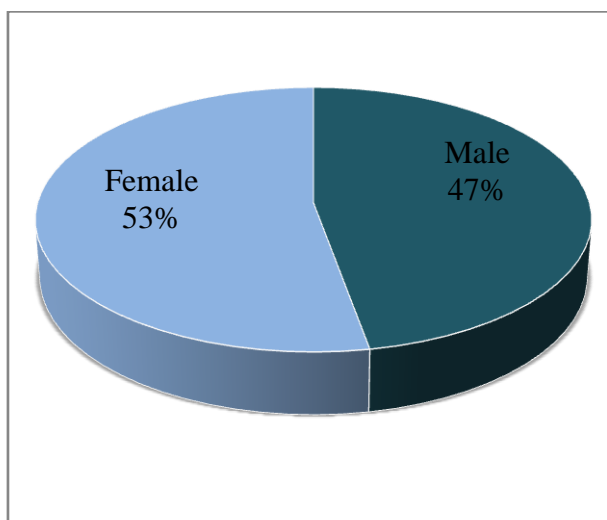


FIGURE 2: Survey by Sex, age group, approximation of years of living near mining areas
 Source: Field Survey, 2020

3.1.3 Classification of Respondents by Sex

The survey results as depicted by the figure 3 about classification of respondent. The survey results as depicted by the about classification of respondents by sex showed that 53% of them were female while only 47% were female.

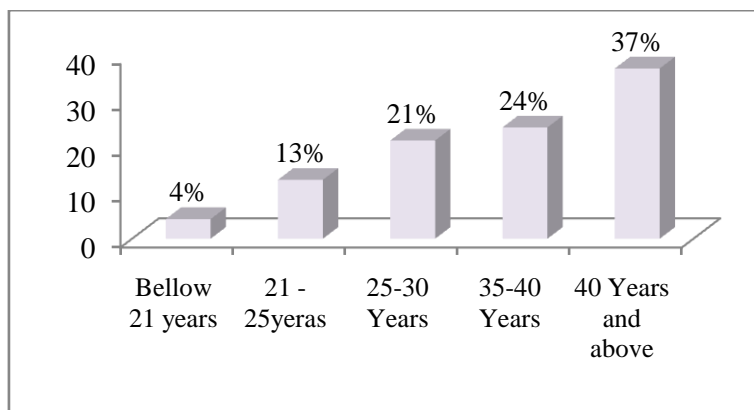


FIGURE 3: Classification of Respondents by Sex
 Source: Field Survey, 2020

3.1.4 Classification of Respondents by Age group

The results of study as portrayed by the figure 4 about classification of respondents by age group indicated that the majority around 37% were aged of 40 years and above, this category is followed by 35 to 40 years, 25 to 30 years, 21 to 25 years and finally the small number of respondents around 4% were aged below 21 years old.

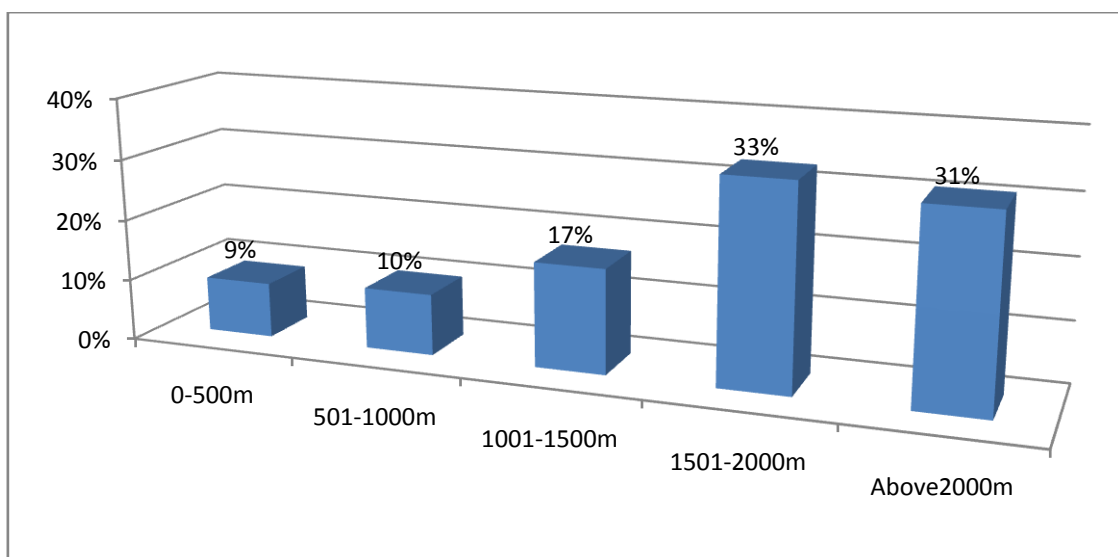


FIGURE 4: Classification of Respondents by Age group
 Source: Field Survey, 2020

3.1.5 Approximate distance near mining areas

The illustration of the figure 5 about approximate distance near mining areas showed that the majority of respondents have their habitation with 1501 to 2000m, 31% replied to have their residence above 2000m from near mining areas. Besides these two categories, 36% resides within distance of 1001 to 1500m, 501 to 1000m and below 500m consecutively in ascending way.

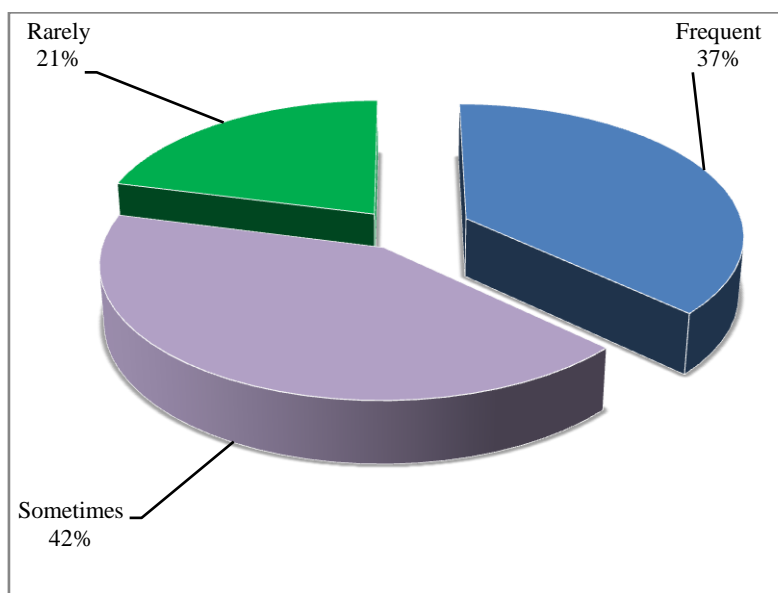


FIGURE 5: Approximate distance near mining areas
 Source: Field Survey, 2020

3.1.6 Frequency of visiting the soil mining area

The 42% representing the majority replied that they sometimes visit the soil mining area; this is followed by 37% who make it happen frequently and last only 21% replied that they rarely visit the soil mining area.

3.2 Impacts of illegal mining on human being in environment

In order to assess the impact of illegal practice on environment, the analysis considered the following: Feeling of Respondents about mineral extraction, Disadvantages of illegal mining from the environment to the residents, to the community, Social, economic and environmental impacts of illegal mining on wellbeing of people.

3.3 Illegal mining practices in Huye District

The results of the survey as depicted by the figure 6 about some of illegal showed that in-situ mining occupies the first position among illegal mining practices that are performed in the selected mining area, this is confirmed by the fact that it holds 41% of total respondents said insitu; 36% of them use surface mining and 14% undertake mining activities using underground mining, whereas (9%) uses placer method.

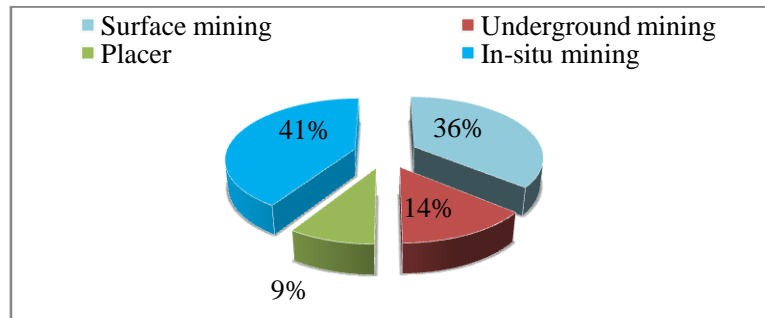


FIGURE 6: Illegal mining practices in Huye District

Source: Field Survey, 2020

3.3.1 Feeling of Respondents about mineral extraction

Most of the time people in surrounding environment say they are skeptical of the quality of the mines, although they often say they have nothing to do with it. They also say that mines often kill people and punish them, but that often ends verbally because it does not prevent miners from continuing to work. For this case, respondents wishes to have owners of mining zone to fast think about the human being in the surrounding area. They suggest things like early expropriation before starting the operations, assuring of people in the surrounding area, establishment of security staff for daily assurance.

3.3.2 Disadvantages of illegal mining from the environment

Residents used said that in addition to the loss of life, illegal mining is damages to the environment through creation of steep slope, leaving workplace with large and open holes that put life of people in a very high risk zone, environmental degradation through water pollution, removal of erosion preventive vegetation, without forgetting the act of shaking house in the surroundings in a very harmful way.

3.3.3 Social, economic and environmental impacts of illegal mining on wellbeing of people

In order to successfully complete the analysis of this survey, it is obvious and compulsory to take a look at the description of social, economic and environmental impacts and then after regression analysis for the test of the claim.

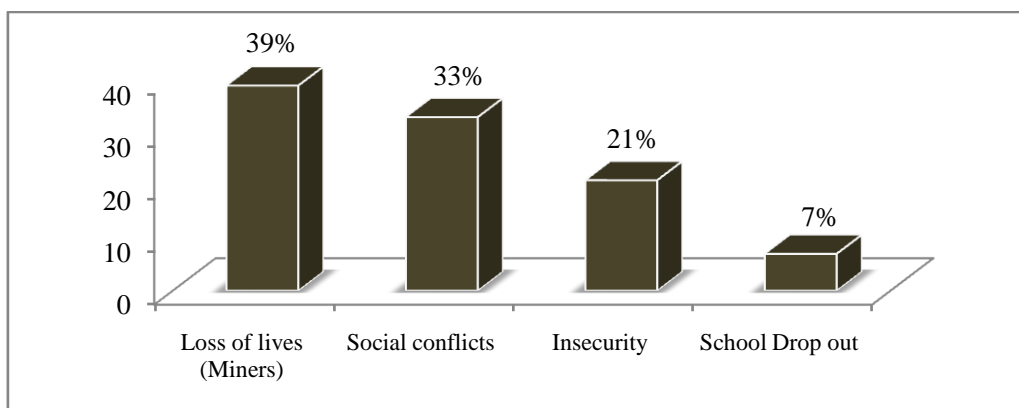


FIGURE 7: Social, economic and environmental impacts of illegal mining

Source: Field Survey, 2020

The perception of respondents as described in the figure 7 about most present social impacts of illegal mining on wellbeing of people indicates that loss of lives occupies the biggest part (39%), this is followed by social conflicts (33%) since the miners fight each other due to the mines they want to accumulate; leading to the several deaths among themselves and around 28% replied the existence of insecurity in the surrounding area and school dropout.

3.3.4 Economic impacts

Normally every activity should provide economic impacts at the end of the day and for this study as it is about illegal mining practices; the analysis is referred to negative impacts on economy.

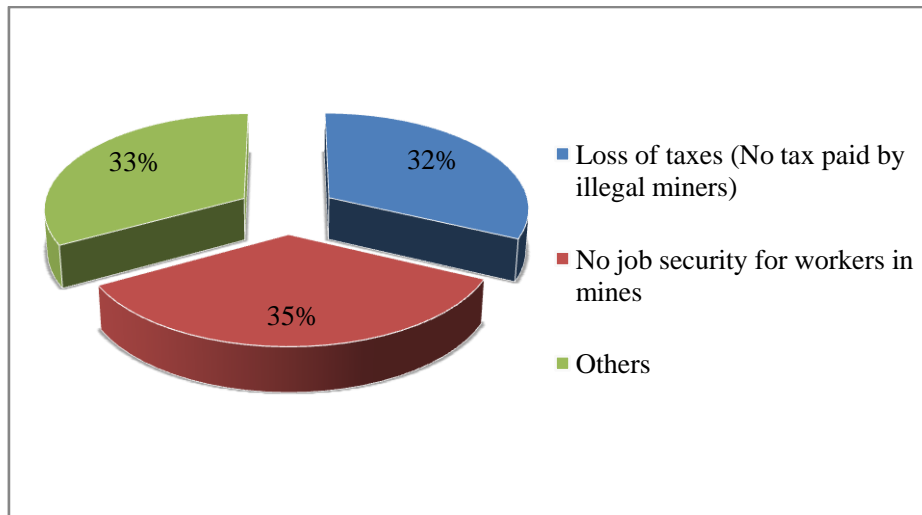


FIGURE 8: Economic impacts of illegal mining

Source: Field Survey, 2020

The results of the study as highlighted by the figure 8 proves that the main negative impact resides on the three impacts with almost equal proportions: Loss of taxes since the illegal miners are not registered as tax payers and this has been confirmed by 32% of the surveyed respondents, 35% confirmed the lack of occupational safety and health during illegal mining while 33% found others impact like careless of their lives and the properties of people living in the surrounding areas.

3.3.5 Environmental impacts

Environmental aspects are elements of an organization’s activities, products, or services that can interact with the environment. In this survey the following are the main negatives of miners on environment: Deforestation, Water pollution, Land degradation/soil erosion and some others although they might be many others not related to this study including but not limited to the following: Hazardous waste , Radioactive waste , Mixed waste , Permitted air emissions, Regulated liquid discharges, Storage, use, and transportation of chemicals , Storage, use, and transportation of radioactive materials, Nonrenewable Energy Use/Intensity (Electricity) and Greenhouse gas emissions.

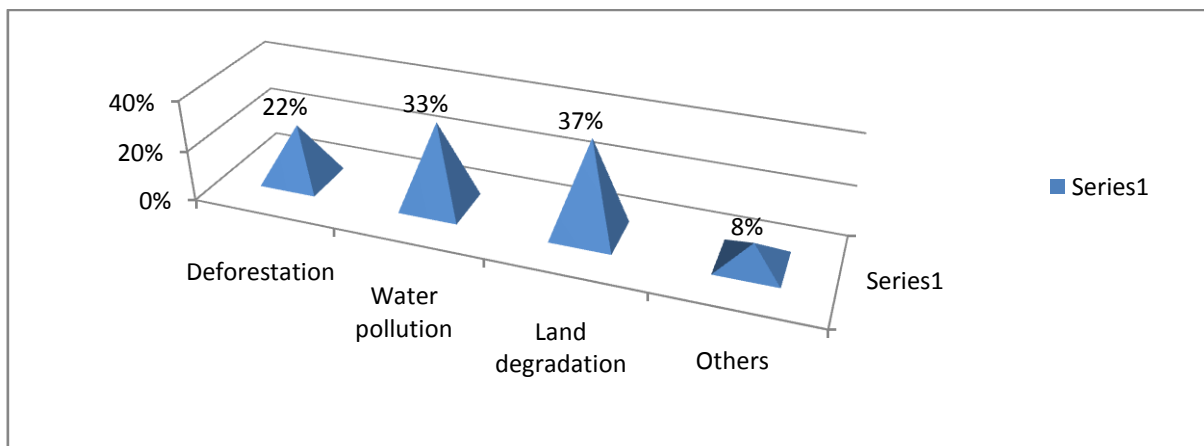


FIGURE 9: Environmental impacts of illegal mining

Source: Field Survey, 2020

The analysis in figure 9 of the data collected among respondents in the surrounding area in which illegal mining took place shows that most and cumbersome problem to resolve locally is land degradation replied by 37% of the surveyed respondents, 33% said the water pollution, 22% make deforestation while only 8% see some other problems.

3.4 Awareness and Reactions of respondents about accident during illegal mining

Hundred percent of respondents have heard the existence of accident during illegal mining through one or more than two of the following channels: verbal communication in the community, radio and social media. Since they are terrible things, most of respondents have had horror reaction.

3.5 Solutions to social, economic and environmental impacts of illegal mining on environment and well-being of people of Huye District, Rwanda

Respondents were asked to show their perceptions on the solutions to social, economic and environmental impacts of illegal mining on environment and wellbeing of people of Huye District; and this is because a lot of negative impacts occurred harming the lives of human being in different ways. However the table below indicates their views.

TABLE 2
DISTRIBUTION OF RESPONDENTS ON SOLUTIONS TO SOCIAL, ECONOMIC AND ENVIRONMENTAL IMPACTS OF ILLEGAL MINING ON ENVIRONMENT AND WELLBEING OF PEOPLE OF HUYE DISTRICT.

Statements	Frequency	Percentage (%)
Public awareness by RMB agents in collaboration with local authorities	98	57.99
Intervention of security organs to prevent illegal mining	21	12.42
Attracting foreign investors by RMB to carry out correctly mining	50	29.59
Total	169	100

Source: Primary data, 2019

The table 2 indicates the perceptions of the respondents on the solutions to social, economic and environmental impacts of illegal mining on environment and well-being of people. A close look to the table shows that 57.99% of the total respondents agree with the Public awareness by RMB (Rwanda Mine, Petroleum and Gas Board) agents in collaboration with local authorities; and it was followed by 29.59% who said that RMB has to attract foreign investors to carry out correctly mining by respecting the rules and regulations governing mining activities in Rwanda. Additionally, the respondents representing 12.4 % of the total respondents approved the intervention of security organs to prevent illegal mining as a solution to the illegal mining and its associated impacts; this is for capturing and punishing those who carry out illegally mining activities and this will surely prevent all negative impacts that occur due to illegal miners; thereby the living conditions of people living around mining areas will be improved.

3.6 Discussion

The current study was about the impacts of illegal mining on wellbeing of people. More specifically the study intended to (i) identify illegal mining practices in Huye District; (ii) assess the social, economic and environmental impacts of illegal mining on wellbeing of people of Huye District; and (iii) find out solutions to social, economic and environmental impacts of illegal mining on environment and wellbeing of people of Huye District.

About the identification of illegal mining practices in Huye District; the study findings indicate that in-situ as the first mining practice was confirmed by 41% of the total respondents; surface mining was approved by 36%; while underground and placer mining practices were revealed and confirmed by 14% and (9%) of the total surveyed population respectively. A close look to these findings and reviewed literatures, one can note that findings of Oladiji et al (2010) reveals four main mining methods of illegal mining such as underground, open surface (pit), placer, and in-situ mining; and according to Albert et al (2015), each mining method has varying degrees of impact on the surrounding landscape and environment.

Concerning to the assessment of social, economic and environmental impacts of illegal mining on wellbeing of people of Huye District, the study depicts the truth. Socially, loss of lives notably of miners was mentioned by (39%) of the total respondents; social conflicts approved by (33%); and insecurity and school dropout were cited and confirmed by 28% of the total respondents. Economically, the study highlighted the loss of taxes to the government due to the fact that illegal miners do not pay tax as confirmed by 32% of the total respondents; lack of occupational safety and health during illegal mining was approved by the 35 % while 33% found others impact like careless of their lives and the properties of people living in the surrounding areas (figure 8). Environmentally, the analysis indicates land degradation replied by 37% of the surveyed respondents; water pollution as approved by (33%); deforestation and other negative impacts of illegal mining were pointed by 22% and 8% respectively. These findings are closely related to that of Abert and Obed (2015) who conducted their study on Environmental Impacts of Mining in Ghana and reported that illegal mineral exploitation contributes significantly to country's GDP, but it is associated with the serious and varying environmental impacts harming to the human lives. Additionally, Joseph (2008) himself through his study on "Environmental and Health Impact of Mining on Surrounding Communities: A Case Study of Anglogold Ashanti in Obuasi" reported that illegal mining leads to the environmental and health impacts to the surrounding communities; notably land degradation, water pollution, air and noise pollution.

IV. CONCLUSION

Basing to the third research objective aiming to finding out solutions to social, economic and environmental impacts of illegal mining on environment and wellbeing of people of Huye District; the study reveals the Public awareness by RMB (Rwanda Mining Board) agents in collaboration with local authorities was pointed by 57.99% of all respondents intervened in the research process; attracting local and foreign investors to carry out correctly mining was said by 29.59% whereas intervention of security organs to prevent illegal mining was a concern for 12.4 % of the total respondents. For this, Tariro (2013) recommended a high level decision making forum involving all stakeholders to discuss problems of illegal mining and how to limit negative impacts.

An analysis done on the Hypothesis Testing through modeling and regression of coefficients, says that $R = 0.809$ representing a very high correlation; and the study was immediately said to have model prediction of the existence Economic, Social and Environmental impacts on human wellbeing, thereby adoption of the null hypothesis. Additionally the existence of the correlation was found statistically significant as its p-value is smaller than 0.05. And this is similar to what Tariro (2013) found in his study "Case Studies of Environmental Impacts of Sand Mining and Gravel Extraction for Urban Development in Gaborone; pit sand and gravel are extracted from open areas creating uncovered deep pits, which caused of accidents to children and livestock, erosion and environmental degradation occur due to continuous mining.

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