ETIOLOGY, INCIDENCE AND MANAGEMENT OF "LEAF YELLOWING SYNDROME" OF RICE (*Oryza sativa* L.) IN MVOMERO DISTRICT

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EXTENDED ABSTRACT

Introduction

Rice (*Oryza sativa* L.) is a source of food and income to smallholder farmers in the Mvomero district. Recently, however, rice farmers in the Mvomero district have been experiencing a new leaf yellowing syndrome of unknown etiology, which is associated with leaf yellowing, affected plant showing stunting reduced tillering and low yield. This study was undertaken with the aim to establish the causal agent(s) of the leaf yellowing syndrome and then develop sustainable measures for its management. Specifically, the study sought to (i) determine the level of farmers' awareness of and management practices for the leaf yellowing syndrome (ii) identify the main causes of leaf yellowing syndrome of organic produced rice in Mvomero District and (iii) develop sustainable management strategies for the leaf yellowing syndrome. The second, third and fourth chapters in the dissertation comprise manuscripts in the form of publishable papers which cover the first, second and third specific objectives.

Methods

With respect to specific objective 1, a field survey was conducted between October – November, 2018 in rice growing areas of Kimambila and Mingo villages in Mvomero District. The aim was to determine the level of famers' awareness of and management practices for rice yellowing syndrome. A face-to-face interview using a semi-structured questionnaire was conducted to gather primary information from 120 randomly selected farmers. Data were analyzed to determine the distribution of respondents per variable using the Statistical Package for Social Sciences (SPSS) program version 16. A Chi-square tests and regression analysis were computed at ($P \leq 0.05$) level of significance, to analyze relationships between variables.

Objective 2; this part of the study was undertaken to identify the cause (s) of the yellowing syndrome in Mvomero district. Rice seeds, leaves and soil samples were collected from the villages under study for assessment. Seed health and seed quality tests were determined using blotter method and agar plate method. Isolation of fungi and biochemical tests of the fungal pathogens were conducted from randomly selected symptomatic rice leaves. The soil samples were analysed for physical and chemical properties following recommended procedures.

Pertaining to objective 3; a study was conducted at Mingo village in Mvomero district, Morogoro region, Tanzania in 2019 aiming at improving rice performance through enhanced soil fertility using organic fertilizers. A split–split plot experiment in a randomized complete block design was laid out with four replications and two factors. The main factor comprised of four rice varieties that included (i) Mbawambili (ii) Mwangaza (iii) Supa and (iv) Saro while the sub factor consisted of (i) Cow dung manure at the rate of 5t ha^{-1}, (ii) Urea 46% N at a rate of 80 kg N ha^{-1} (iii) Compost at the rate of 5t ha^{-1} and (iv) No fertilizer (control). Rice was sown in 2m x 1m plots each with five rice rows at 20cm inter–and intra–row spacing. Data on weather, leaf yellowing incidence, crop growth, yield components and grain yield were collected. Data were subjected to the analysis of variance (ANOVA) using Genstat 16th edition software (VSN International). Means separation test was conducted using Turkey’s Honest Significance Difference Test using $P \leq 0.05$ as the level of significance. A correlation analysis was calculated using Pearson product moment correlation coefficient (r) using the Microsoft office-Excel programme to explore the relationship between growth and yield components of rice.

Findings

Yellowing and stunting of rice were reported as predominant problems by 100% of interviewed farmers. Lack of knowledge on rice leaf yellowing syndrome was reported by all (100%) respondents as the main factor limiting the management of the yellowing syndrome. *Bipolaris oryzae* and *Fusarium moniliforme* pathogens in tested seeds were found to be predominant in all tested rice varieties. Soil pH was found suitable for rice production; Nitrogen and Potassium were very low than the suggested standards and therefore deemed deficient. Nitrogen deficiency in both sites was the major causes of yellowing syndrome reported. Disease index results indicated that all the fertilized rice varieties did not develop yellowing and stunting syndrome. However, high incidence of the yellowing syndrome and plant stunting were observed in the unfertilized control plots implying that N deficiency is the main contributing factor to the leaf yellowing and stunting syndrome. There were significant ($p<0.01$) effect in respect of varieties and fertilizer types used on growth parameters and yields components. Further, regression and correlation analyses showed positive correlation between yield and other growth parameters.

Conclusions and recommendations

Generally, lack of knowledge on the cause of rice leaf yellowing syndrome was the main factor limiting the effective management of the syndrome in Mvomero district.

Several seed borne pathogenic fungi were found associated with rice seed samples from Mingo and Kimambila villages and therefore indicate the possibility of disease episode when such infected seeds are used to raise the next rice crop. Nitrogen deficiency was confirmed to be the major cause of yellowing syndrome. Measures should be taken to increase the availability of macronutrient in the soil especially N and P in order to increase rice productivity. Uses of organic manure together with other sustainable agronomic practice such as fallowing, intercropping, and planting leguminous crops in the field alongside rice varieties will help adjust soil fertility and spar rice yield. Mbawambili which is a commonly used local rice variety does not fair favourably against Supa, Mwangaza and Saro with regard to yield and yellowing syndrome. Therefore a breeding program to incorporate genes for higher yield and disease resistance in the variety should be undertaken in order to increase its disease resistance and yield potential.

DECLARATION

I, Hekima B. Mliga do hereby declare to the Senate of Sokoine University of Agriculture that this dissertation is my own original work done within the period of registration and that it has neither been submitted nor being concurrently submitted for a degree award in any other institution.

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The above declaration is confirmed by

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DEDICATION

I dedicate this work to my parents Mr. and Mrs. Mliga who stand with me through it all and ensuring that I achieve my dreams and provide me with everything I needed. To every girl chasing your dream out there keep on going, work so hard to grab it all and make your society proud.

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LIST OF ABBREVIATIONS

^0C	Degree Centigrade
AAS	Atomic Absorption Spectrophotometer
B	Boron
Ca	Calcium
CEC	Cation Exchange Capacity
Cu	Copper
DAP	Diammonium Phosphate
DAP	Days after Planting
DF	Degree of Freedom
DTPA	Diethylenetriaminepentaacetic acid
EC	Electric conductivity
EUCORD	European Cooperative for Rural Development
FAO	Food and Agriculture Organization
FAOSTAT	Food and Agriculture Organization Statistical
Fe	Iron
GDP	Gross Domestic Product
GI	Germination Index
HI	Harvest index
Ha	Hectare
IITA	International Institute of Tropical Agriculture
IRRI	International Rice Research Institute

ISTA	International Seed Testing Agency
K	Potassium
kg	Kilogram
KOH	Potassium hydroxide
MC	Moisture content
Mg	Magnesium
Mm	Millimeters
Mn	Manganese
N	Nitrogen
Na	Sodium
NPK	Nitrogen, Phosphorus, Potassium
P	Phosphorus
PDA	Potato Dextrose Agar
RH	Relative humidity
RYMV	Rice yellow mottle virus
S	Sulphur
SAT	Sustainable Agriculture Tanzania
SOC	Soil Organic Carbon
SPSS	Statistical Packages for Social Sciences
SUA	Sokoine University Agriculture
T	Tons
TEA	Triethanolamine
TMA	Tanzania Meteorological Agency
TSH	Tanzanian Shilling

URT	United Republic of Tanzania
USDA	United State Department of Agriculture
X^2	Chi Square
Zn	Zinc

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background Information

Rice (*Oryza sativa* L.) is an important food and cash crop cultivated around the world (Mahadevappa, 2004). It is a staple food for over 4.2 billion people, which is more than half of the world population (Amna *et al.*, 2006). It is also the primary source of income and employment for more than 200 million households across countries in the developing world (Muthayya *et al.*, 2014). Rice was introduced in Tanzania in early 1890s and has become an important second staple food after maize, consumed by 60% of the population and grown in almost all regions of the country (FAO, 2015). The smallholder rice farmers depend on the crop for both food security and cash. Mbeya and Morogoro are the main rice producing regions; other regions which significantly produce rice are Arusha, Kigoma Kilimanjaro, Pwani, Rukwa and Shinyanga (Hubert *et al.,* 2016).

Total production of rice in Tanzania during 2016/2017 cropping season was reported to be 1.38M tons. Morogoro was the leading region producing 24 percent of the total rice produced, followed by Mbeya which contributed 17.8 percent, then Tabora 12.4 percent and Njombe which produced 0.02 percent of the total rice produced (URT, 2017). Tanzania is the largest rice producer in East Africa, producing about 80 percent of all rice; also it is the second in Eastern, Central and Southern Africa in terms of rice production and consumption after Madagascar (Rugumamu, 2014).

Despite the high demand for rice worldwide and Africa in particular, the productivity of the crop has remained low in most rice producing countries of Africa continent including Tanzania. Rice yield on smallholders fields in Tanzania range from 1.0 - 2.2 tons ha^{-1} compared to the yield of 5.3 - 6.0 t ha^{-1} and 5.0 t ha^{-1} reported in the rice-growing countries of Vietnam and China, respectively (FAO, 2015). Thus, there is a potential for increasing rice productivity in Tanzania.

A number of biotic and abiotic stresses have been reported to contribute to the low yield of rice in Africa of which poor soil fertility, low yielding cultivars, insect pests, and diseases are the most important (Lamo *et al.*, 2015; Kalala *et al.*, 2016). Rice yellow mottle virus (RYMV), rice blast (BL), and bacterial leaf blight (BLB) are the most significant diseases affecting rice productivity in Tanzania and can cause up to 100% yield losses(Luzi-Kihupi *et al.*,2015).

Other factors that contribute to decrease in rice yield are inadequate rainfall and long term continuous utilization of soil without applying balanced nutrients. Soil fertilization is one of the major inputs which contribute to increase in yield and quantity of rice. However, nutrients uptake from the soil by the crops may vary from one field to another due to plant or soil factors such as texture, organic matter content, structure, root depth and soil depth (Janaki, 2006). Several studies have shown that disease incidence is influenced by a number of factors including genetic background of the host plant, vector population and climatic conditions.

1.2 Leaf Yellowing Diseases of Rice

1.2.1 History and geographical distribution

Rice yellow dwarf disease (RYDD) has been reported to be a problem for rice farmers in many rice-producing regions of Asia (Jung *et al.,* 2018). Infected rice plant turns pale yellow and gradually starts to decay; it ultimately shows stunted growth and fails to produce grain (Fig. 1.1). For many years after its discovery in 1919, the agent that caused this disease was unknown and was believed to be a virus until it was identified as a Phytoplasma then called a mycoplasma-like organism (Jung *et al.,* 2018). There are no reports on the existence of Rice yellowing dwarf disease (RYDD) or Rice yellow stunt disease (RYSD) in Tanzania although a disease with symptoms nearly similar to RYDD and RYSD have been observed in fields of rice grown under sustainable agriculture in Mvomero district recently (Mliga 2019, Personal observations).

Figure 1.1: Plants showing symptoms of yellowing Syndrome in Kimambila village, Mvomero 2018

1.2.2 Symptoms of “Rice leaf yellowing disease”.

The symptoms of “Rice leaf yellowing disease” (RLYD) as observed on susceptible rice cultivars in Mvomero District are leaf yellowing, plant stunting, narrowing of emerging leaves, incomplete panicle emergence, plant wilting with subsequent black colouring of the plant and, severe yield reduction. These symptoms closely relate with those of RYMV, Rice orange leaf disease (ROLD), RYDD and Rice yellow stunt disease (RYSD). However, multiple infections can also result into symptoms akin to those of RLYD as reported for co-infection of bacteria and viruses or nutrient deficiency diseases (Luzi-Kihupi *et al.*, 2000; Phuongl *et al.*, 2011).

Symptoms of Rice Tungro disease and Rice Yellow Stunt Disease resemble those of ROLD, with the leaves becoming yellow along the leaf vein, stunting of plants, reduced tillering and poor root development (Li *et al.,* 2015). Rice yellow dwarf (RYD) and Rice orange leaf (ROL) are the two Phytoplasma diseases that have been reported to infect rice. RYD, a serious problem for rice farmers, has only been detected to date in Asia, where it has been recorded from most rice-growing countries (Valarmathi *et al.,* 2013). Infected rice turns pale yellow and gradually starts to decay and produce numerous tillers.

Pham *et al.* (2007) reported rice plants showing symptoms of “yellowing” and “tungro-like symptom” in the Mekong Delta in 1989, and the disorder became a serious problem after 1994. Affected rice plants showed light spreading growth habit, light stunting and yellowing or light yellow to dark orange discoloration of the leaves. Sometimes only few tillers per hill appeared infected, without profuse tillering or further growth.

1.2.3 Epidemiology of rice leaf yellowing syndrome

The epidemiological characteristic of the leaf yellowing syndrome presumably indicates the involvement of biotic factors such as viruses and insect vectors, although the exact causes of the disease cannot clearly well elucidated based only on symptoms. Thus, the underlying causes of rice yellowing disease epidemic in Mvomero might have been incited by one or more of the above mentioned factors but may also be due to changing cultural practices, nutrients deficiencies, crop monoculture and obvious mechanical damage (Phuongl *et al.*, 2011). However, the exact cause of the rice leaf yellowing disease in Mvomero could only be ascertained after a careful diagnosis of the causal agent(s) involved.

1.2.4 Isolation and detection methods of rice diseases

Methods for detection of seed borne microorganisms are ranged from simple visual observation to spectroscopic and imaging techniques. They vary depending on the pathogen in question. The most common seed health tests including washing test (WT), blotter method (BM), washing after incubation, modified (WAI), deep freezing method (DF) and potato dextrose agar method (PDA). The advanced techniques are DNA extraction technology and Nano-PCR which is quite new area in the field of biotechnology (El-Abbasi *et al.*, 2020).

1.2.5 Economic importance of rice leaf yellowing syndrome in Mvomero District, Tanzania

Yield losses caused by RYD on rice range from 20 – 100% depending on plant age prior to infection, susceptibility of the rice variety and environmental factors such as rainfall, humidity and temperature (Kouassi *et al.,* 2005b; Salaudeen, 2014). Li *et al.* (2015) reported loss of about 31 000 ha of rice fields due to infection of ROLD. According to farmers' testimony, the leaf yellowing disease of rice in Mvomero district reduces yield by up to 40%, which is very high. However, until now no research has been done to ascertain the farmers' claim. There is, therefore, a need for a scientific research to elucidate the exact impact of the disease on the rice crop.

1.2.6 Management of leaf yellowing syndrome

The management strategies for the yellowing disease will initially involve screening of local and imported rice germplasm for resistance against the disease, other practices will include chemical treatment, plant quarantine, development of efficient cultural practices, and integrated disease management strategies (Abo and Sy 1997; Phuongl *et al.,* 2011).

Integrated management package including cultural control methods, general sanitation, use of recommended plant spacing, timely planting and avoiding contamination during land preparation and field management in order to decrease the inoculum pressure (Uke *et al.,* 2014). The specific strategies to be used to manage the disease, however, will be developed after the cause of the disease has thoroughly been identified.

1.4 Problem Statement and Justification

Rice is affected by many diseases caused by fungi, bacteria, viruses, nematodes, Mycoplasma and nutrients deficiencies (Lamo *et al.*, 2015). The incidence of abiotic and biotic diseases and the damage they cause to rice crops are reported to be increasing wherever the crop is grown (Zouzou *et al.*, 2008). An integrated approach to crop management including host plant resistant, appropriate control of plant pathogens and mitigation of soil nutrients could reduce much of the disease problem. However, in developing countries, such a strategy is rarely applied effectively. The main reasons for the non- effective application of the control measures are the apparent lack of adequate knowledge of aetiology and incidence of the diseases.

It was recently reported by rice farmers from Mvomero district during the 4^{th} Sustainable Agriculture Tanzania workshop in Morogoro that for the past four years they have been experiencing a new problem of rice which is associated with leaf yellowing, plant stunting, reduced tillering and low yield (SAT, 2017). The cause of yellowing and stunting of rice in Mvomero is not known. However, the involvement of either biotic plant pathogens or nutrients deficiencies in the soil cannot be discounted.

Although no yield loss studies have been conducted yet, yield reduction of up to 40% has been reported by the organic rice farmers in Mvomero district in recent years (SAT, 2017). Given such situation, this syndrome not only poses a serious threat to the most important food crop in the country but also the livelihood of the people who depend on the crop as a source of income, food, employment and food security (FAO, 2015).

Research information on factors causing low yield of rice under sustainable agriculture production system in Tanzania are unavailable. This study, therefore, sought to identify the biotic, abiotic, social or economic factors causing leaf yellowing and yield reduction in rice and their impact under sustainable agriculture in order to develop effective strategies for increasing rice productivity and income of small scale farmers in Mvomero district.

1.5 Research Objectives

1.5.1 Overall objective

Establish the main cause(s) of leaf yellowing and low yield of rice in Mvomero district with a view to developing sustainable strategies for their management.

1.5.2 Specific objectives

i. To determine farmers' awareness of and management practices for rice leaf yellowing syndrome in smallholder fields in Mvomero district.

ii. To identify main causes of rice leaf yellowing and low yield of organic rice in Mvomero district.
iii. To develop sustainable management strategies for the leaf yellowing syndrome of rice in Mvomero district.

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CHAPTER TWO

2.0 KNOWLEDGE LEVEL AND MANAGEMENT PRACTICES FOR LEAF YELLOWING SYNDROME AMONG SMALLHOLDER FARMERS

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2.1 Abstract

A field survey was conducted in rice growing areas of Kimambila and Mingo villages in Mvomero District between October – November 2018 to determine the level of farmers' knowledge of a new rice leaf yellowing syndrome and management practises against the disease. The two villages were purposively selected for the survey based on the report of the occurrence of leaf yellowing syndrome and their long history of rice farming under sustainable agriculture. A face-to-face interview using a semi-structured questionnaire was conducted in the two villages to gather primary data from 60 farmers in each village. Respondents were selected using a multi-stage random sampling procedure. Yellowing and stunting of rice were reported as predominant problems by 100% of the farmers. Most (90%) of the farmers had experienced the syndrome for a period of less than one year prior to the survey; a few (10%) had seen the disease for between one to three years. Some (60%) farmers associated the disease with a type of rice variety currently in production; others (40%) believed drought and witchcraft were the main causes. Most (55%) farmers reported Supa as the variety most susceptible to the disease. In both villages 100% of famers do not use any measures to control the disease, while 80% use weeding and mixed cropping as control measures. Lack of knowledge of and control measures for the disease are undoubtedly the main factors limiting the management of the new rice disease in famers' fields in Mvomero district. To improve rice yield, it is hereby suggested that a thorough and accurate identification of the main cause(s) of the syndrome is necessary before any effective management strategies can be developed.

Key words: Mvomero District, *Oryza sativa*, Yellowing and stunting syndrome.

2.2 INTRODUCTION

Rice (*Oryza sativa* L.) is the most widely grown crop and a staple food worldwide. The crop is cultivated in a variety of water regimes and soil types (Muthayya *et al.*, 2014). It is the primary staple food for more than 51% of the World's population (January, 2012). People depend on rice for food calories and protein, and more than one fifth of the world's population or more than a billion household in Asia, Africa and the America rely on rice systems for their main sources of employment and livelihoods. It is also on the forefront in the fight against food insecurity, hunger and poverty (Nguyen and Ferrero, 2006). Over 90% of the world's total rice crop is produced in South and East Asia. China is the leading rice producing country in the world producing up to 148.5 million metric tons. In the year 2017/2018 total area harvested rice worldwide was 167.13M ha and total yield production was 782.0M tonnes. Africa contributes 4.2% in the world total rice production (FAOSTAT, 2018). Rice is becoming an increasingly popular food in Africa because it is easy to store, cook, tasty and can be used for a large variety of dishes. It is grown in more than 75% of African countries, with a combined population of close to 800 million people. Tanzania is the second largest producer of rice in East Africa, after Madagascar. According to FAOSTAT (2018) in 2017/2018 Tanzania produced 2.9 M tonnes of rice (Table 2.1). Rice ranks second as staple and cash crop after maize; however its productivity is low and it is continuously declining (Kalala *et al.*, 2016). EUCORD (2012) reported that the rice sector is among the major sources of employment, income and food security for Tanzanian farming households; and it ensures a staple food supply for the urban population. An estimated 18% of farming households grow rice, and the economic activity in the rice sector contributes to 2.66% of the national GDP. With steadily growing demand due to increases in per capita consumption and population growth, the total area under rice cultivation has also increased substantially.

Rice consumption in East Africa Countries region stood at 2.844M, MT in 2015 and increased consumption is attributed to various factors such as population growth, urbanization, change consumers preference and economic development (Kilimo Trust, 2017).

Table 2.1: Tanzania rice production statistics for the years 2013-2018

Year	Area harvested (ha)	Production (tonnes)	Yield (kg/ha)
2013	928 273	2.19M	23 643
2014	957 218	2.6 M	27 382
2015	1 154 467	2.9M	25 812
2016	1 241 722	3.0M	24 312
2017	1 165 238	2.8M	24 609
2018	1 199 875	3.0M	25 142

Source: FAOSTAT (2018)

About 71% of the rice grown in Tanzania is produced under rain fed conditions, where irrigated land presents 29% of the total land with most of it in small village level traditional irrigations with the average yield of 1-1.5 t/ha (Kibanda, 2008). Various factors have been highlighted as the major cause of decrease in productivity of rice; these factors include pests, droughts, low soil fertility, over utilization of soil without fertilization, nutrient imbalance and poor local seed varieties (Agritrade, 2012; Kalala *et al.*, 2016).

Recently, rice farmers at Kimambila and Mingo villages reported that their rice crop is often yellow and stunted and they obtain very low yield (SAT, 2017). Based on visual examination of the diseased plants in the field it was presumed that the problem was either due to biotic or abiotic causes. Therefore a survey to determine the level of knowledge of and management practices for leaf yellowing syndrome and low yield among smallholder farmers was deemed necessary to obtain detailed information on the problem and then use the obtained information to develop a sustainable management strategy.

2.3 Materials and Methods

2.3.1 Description of the study site

The study was conducted at the villages of Kimambila and Mingo in Mvomero district of Morogoro Region from October to November, 2018 (Fig. 1.1). Mvomero district is located at Latitude: 6°18′0″ South, Longitude 38° 41' 37.4928" East and elevation of 400-1500 meters above sea level. The average annual rainfall in this district is 1000 mm while the temperatures vary depending on the season, but generally the average minimum and maximum temperature is 20 and 30ºC respectively.

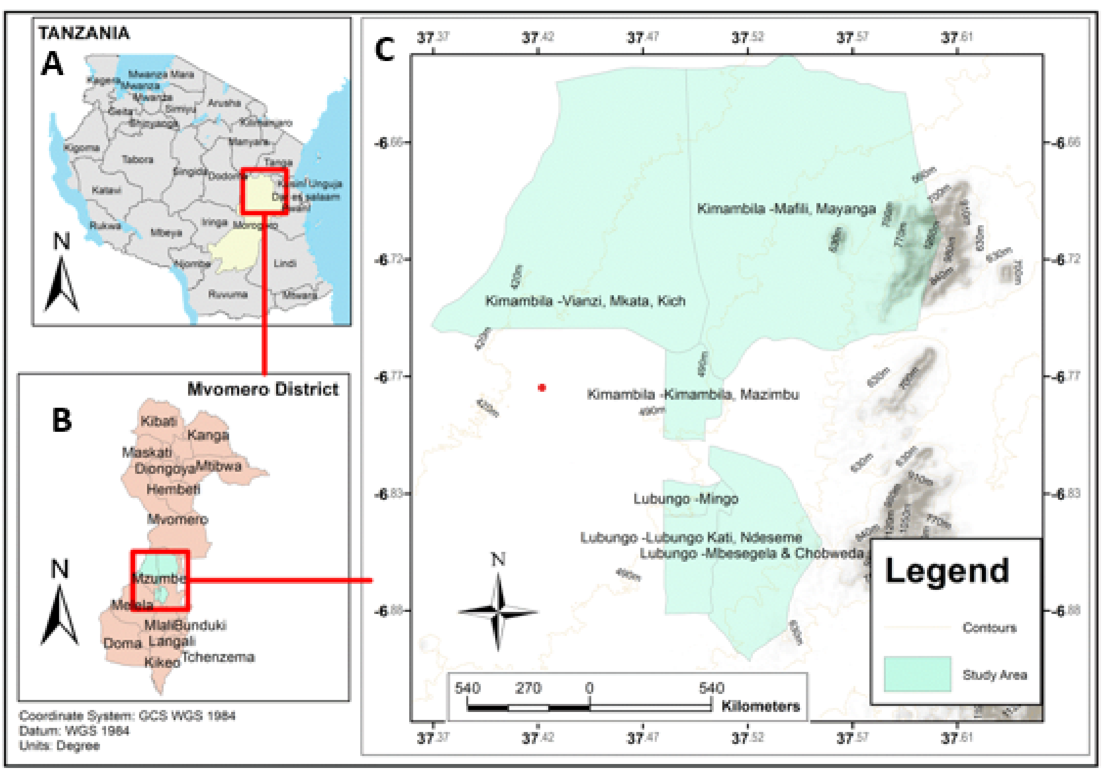

Figure 2.1: A. Tanzania. B. Mvomero district. C. Surveyed villages in the study area

2.3.2 Sample Selection

Respondents were selected using a multi-stage random sampling procedure as per Schreinemachers *et al*. (2015). Mvomero district was selected for the survey based on its long history of rice production and previous report by farmers on the presence of leaf yellowing syndrome in their fields (SAT, 2017). From the district, two villages Mingo and Kimambila were purposively selected for the survey. The villages were selected based on the availability of farmers groups growing rice under the direction of SAT organization. The sample size (n) i.e. the number of farmers for the interview was determined as per Wonnacott and Wonnacott (1990) as cited by Mghase *et al*. (2010) using the following equation.

$$N = \frac{Z^2 P(1-P)}{Q^2} = \frac{1.96^2(1-0.9)}{0.05^2} = 138 \qquad (1)$$

Where, N = required sample size, Z = confidence level at 95% (standard value of 1.96), p = estimated proportion of an attribute (average percent of Rice farmers in a population of farmers in the villages), estimated at 90% and the Q = margin of error at 5% (standard value of 0.05). Thus one hundred thirty eight (138) farmers were selected for interview but due to limitation of resources and unavailability of enough farmers groups in the villages the sample size was hence reduced to 120 making a sample size of sixty respondents from each village. Sixty of 120 farmers interviewed were women and the rest were men.

2.3.3 Data collection

2.3.3.1 Survey data

Data were collected through face-to-face interviews and filled in a semi-structured questionnaire (Appendices 1). The questionnaire was composed of questions related to farmers' preferences in type of rice varieties grown, their current management practices for the leaf yellowing syndrome and whether the farmers' were aware of the prevailing syndrome problem. The data on farmers' demographics profiles (e.g. age, sex, gender, and education), agronomic practices performed on the crop and farmers experience on rice farming were also collected.

2.3.4 Data analysis

Quantitative and qualitative data were coded and subjected to statistical analysis using the Statistical Package for Social Sciences software (IBM SPSS Statistics 20). Cross-tabulations tables were constructed and descriptive statistics were calculated to summarize data from the questionnaires. To make statistical inferences, contingency chi-square tests were computed at $P \leq 0.05$ level of significance to analyze relationships between variables.

2.4 Results

2.4.1 Demographic characteristics

The demographic characteristics of interviewed farmers are presented in (Table 2.2). Among the interviewed farmers, 50% were males and 50% were females. The farmers' age ranged from 19 to 63 years. When asked about their level of education, 15% of the respondents had no any formal education, 70% had primary school education and only 15% had secondary school education.

Table 2.2: Demographic characteristics of farmers in the surveyed area (n = 120)

Characteristic	Kimambila	Mingo	Mean (%)	Df	χ^2	*p*-value
Age (Years)						
18-35	14.3	23.1	20			
36-45	42.9	53.8	50	2	5.275	0.072
>46	42.9	23.1	30			
Sex						
Male	57.1	46.2	50	1	1.319	0.251
Female	42.9	53.8	50			
Education level						
No formal education	14.3	15.4	15			
Primary	71.4	69.2	70	2	0.063	0.969
Secondary	14.3	15.4	15			

Note. df = degrees of freedom, χ^2 = Chi-Square test, $p \leq 0.05$ shows there was a significant difference.

2.4.2 Farm characteristics

There was a significant ($P<0.05$) difference between the surveyed villages on the number of farmers with experience in farming, sizes of their farm, rice cropping systems and the type of minor crops grown alongside rice (Table 2.3). At Mingo village, 69.2% of farmers had more than 5 years' experience in rice production. Majority (60%) of famers at Mingo village own more than 1.1 ha of land and practiced sole cropping. At Kimambila village, 71.4% of interviewed famers had experience of less than one year in rice production and 71.4% practice mixed cropping with rice as the main crop and maize and sunflower as minor crops.

The most common cropping systems practiced by farmers were mixed and sole cropping systems. Most farmers (60%) cultivated rice only as major crop, 40% cultivated rice and maize.

Table 2.3: Characteristics of the surveyed farms (n = 120)

Farm characteristic	**Kimambila**	**Mingo**	**Mean (%)**	**df**	**χ^2**	***P*-value**
Farm experience (Years)						
<1	71.4	0	25	2	84.84	<0.01
2-4	28.6	30.8	30			
>5	0	69.2	45			
Farm size (Ha)						
<1	71.4	23.1	40	1	26.59	<0.01
>1.1	28.6	76.9	60			
Cropping systems						
Mixed cropping	71.4	38.5	50	1	11.87	<0.01
Sole cropping	28.6	61.5	50			
Type of minor crop						
Maize	42.9	38.5	40	2	28.35	<0.01
Sunflower	28.6	0	10			
No minor crop	28.6	61.5	50			

Note. df = degrees of freedom, χ^2 = Chi-Square test, P ≤ 0.05 shows there was a significant difference.

2.4.3 Rice production and economic benefits

Table 2.4 shows most (50%) farmers harvested 0.25 – 0.5 t/ ha of rice during 2017/18 season. About 55% farmers sold their rice at a price ranging from 700 – 800 Tshs/kg, while 45% sold at price of between 900 –1000Tshs/kg. There was a significant difference (P<0.05) across the surveyed villages on rice yield. At Mingo village, 38.5% of farmers harvested 0.76 – 1.02 tons of rice / ha and most sold their rice at a price ranging from 900 – 1000 Tshs/kg. At Kimambila village, most (71.5%) of farmers harvested 0.25 – 0.5 tons of rice / ha and sold for 700 – 800 Tshs/ kg.

Table 2.4: Rice production and economic benefits (n = 120)

Rice production	Kimambila	Mingo	Mean (%)	df	χ^2	*P*-value
Rice contribution to income						
Moderate	71.4	92.3	85	1	9.33	<0.01
Low	28.6	7.7	15			
Rice yield (tons/ha)						
0.25-0.5	71.4	38.5	50	2	22.42	<0.01
0.51-0.75	28.6	21.3	25			
0.76-1.02	0	38.5	25			
Rice price (Tshs/kg)						
700-800	85.7	38.5	55	1	24.63	<0.01
900-1000	14.3	61.5	45			

Note. df = degrees of freedom, χ^2 = Chi-Square test, $p \leq 0.05$ shows there was a significant difference.

2.4.4 Constraints of rice production

Yellowing and stunting of rice were mentioned as predominant production problems among rice farmers in both villages (Fig. 2.2). All interviewed farmers associated yellowing and stunting of rice with the type of rice variety cultivated. There was a significant difference (P<0.05) across surveyed villages on the possible cause of yellowing and stunting of rice. At Mingo village, 46.2% of respondents reported drought as the main cause of yellowing and stunting while witchcraft was reported by 47% farmers at Kimambila village.

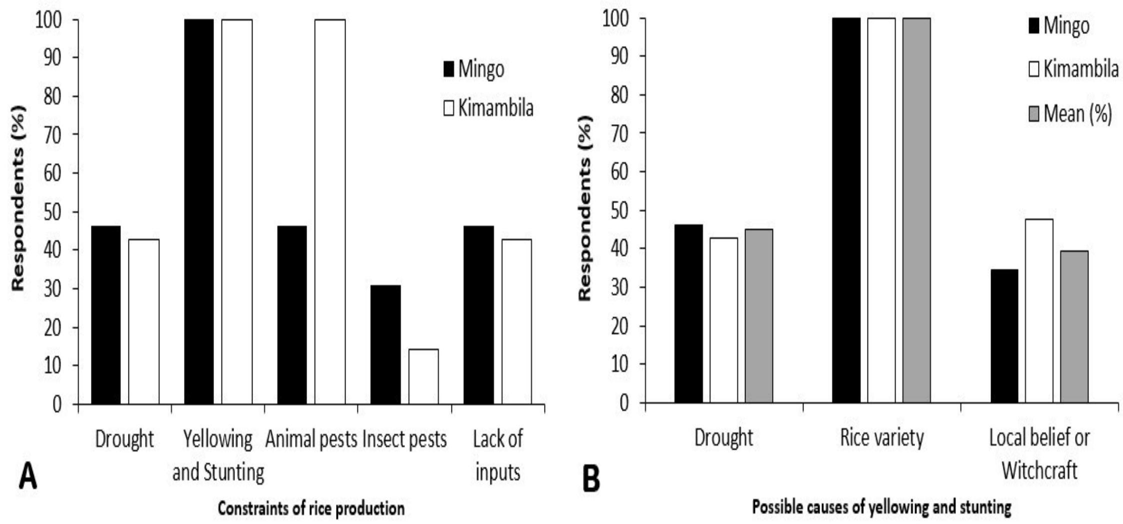

Figure 2.2: A. Farmers response on the constraints of rice production. B. Farmers' perception about possible causes of yellowing and stunting problems

2.4.5 Occurrence of rice yellowing and stunting problems

Most (90%) farmers have experienced the rice yellowing and stunting problem for a period of less than one year (Fig. 2.3). There was a significant difference ($P<0.05$) between surveyed villages on how long the farmers had experienced the yellowing problem and type of susceptible varieties cultivated. Majority (90%) of farmers at Mingo village had experienced the problem for a period of between one and three years, while all farmers at Kimambila village had experienced the problem for less than one year. Regarding rice varieties most susceptible to yellowing syndrome, most of farmers reported Supa (55%) and Mbawambili (45%) as the most susceptible varieties.

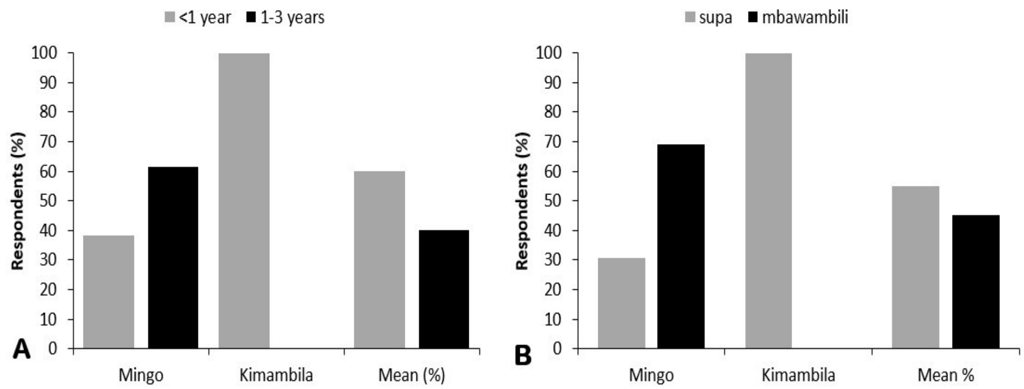

Figure 2.3: A. Farmers response on the period/ time experienced rice yellowing and stunting. B. Rice varieties susceptible to yellowing and stunting

2.4.6 Management of yellowing and stunting syndrome

All 100% of respondents in both villages do not use any disease management method; however all 100% of respondents reported weeding as the main practice in their rice fields (Fig. 2.4). Lack of knowledge on control methods was reported to affect the management of the syndrome by all respondents.

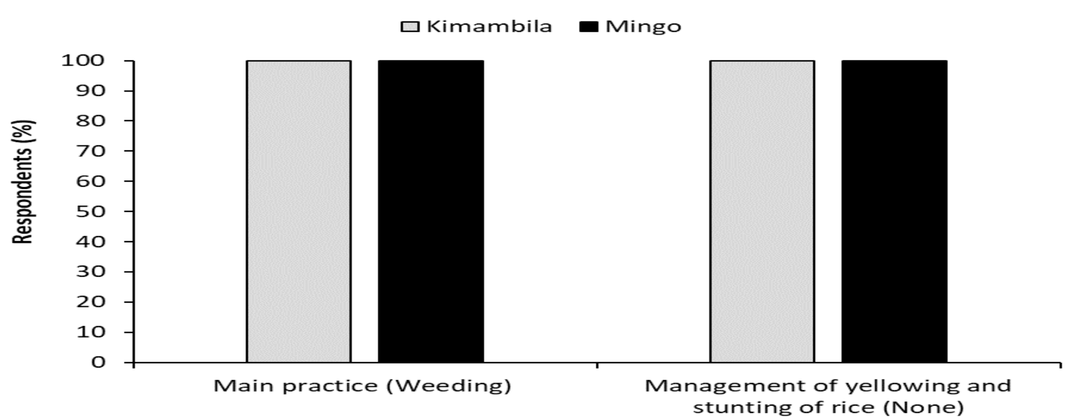

Figure 2.4: Farmers responses on the main agronomic practices performed on the rice crop and the measures used to combat the rice yellowing and stunting syndrome

2.5 Discussion

2.5.1 Demographic data

Both males and females in the surveyed villages are actively and equally participate in rice farming activities, and this had been reported elsewhere by Hashim *et al.* (2018). In a study of determinants of rice preference in Kilimanjaro, Laizer *et al.* (2018) reported participation of women by 56% in agriculture. In his study, Ngailo *et al.* (2016) reported that majority (89.6%) of the respondents were males while a few (10.4 percent) were females. A majority of Tanzanian farmers are women and make a significant contribution to food production and to the processing and marketing of foodstuffs. They form 60 - 80% of the agricultural labour force in the rural areas (URT, 2009). Although both men and women participated in farm activities, ownership of land and distribution of resources after harvesting are handled by males. These results also correspond to other studies of land ownership and decision making as observed by Msangya and Yihuan (2016).

Distribution of age across respondents was also equal in all villages. Half of respondents (50%) were aged between 36-45years; youths (18-35) years which were 20% of respondents are not actively engaged in farming activities. This phenomenon was clearly observed during interview as most of respondents were adults. Similar results were observed by Laizer *et al.* (2018) where 60% of respondents aged between 36-45 were actively engaged in agriculture. Youths in both villages are more involved in transportation services as drivers of cars and motorcycles and majority of them have migrated to nearby urban areas like Morogoro town and Dar es Salaam.

The same was observed and reported by Muhammad *et al.* (2015) where 87.4% of the respondents belong to middle and older age groups. These results shows that youth are not actively engaged in agriculture and this may threaten production and availability of rice in Mvomero district and at the national level in general. To increase participation of youth in agriculture the government through village agricultural officers should conduct seminars and farmers field schools in the villages and engage them throughout the process. Furthermore provision of equipment of financial support may also encourage participation of youth in the sector. Education and age plays important role in agriculture output because when the youth are educated it become easy to use their knowledge and older men's experience to increase production.

70% of respondents had primary education and a few (15%) of respondents had secondary education while the remaining 15% of respondents had no any formal education. Ngailo *et al.* (2016) in their study reported that majority of the respondents had formal education that could be capitalised for rational decision-making regarding rice production and marketing without difficulty and preceding fact shows that a decision to engage in improved farming was related to the level of education that respondents had. Lack of education might be one of the reasons for poor performance of crops as these farmers use local farming techniques' and they don't have the mettle to adopt new technologies.

From group discussion and observation, farmers' associate low production with witchcraft. Muhamad *et al.* (2015) also reported that illiteracy increase communication barrier especially when new farming concept and techniques are to be introduced. In addition, Amaza *et al*. (1999) found that lack of awareness due to low literacy rate hinders women's access to farm resources in Nigeria and thus affect their productivity.

Results also showed that farmers had sufficient experience in rice farming of more than five years. Most of them are adults and they have practiced farming for long time, farmers from Mingo village are more experienced in rice farming as compared to those in Kimambila village. In a study by Wiredu *et al.* (2010) on rice cultivation in Ghana, age had positive effect on yield meaning experience in rice cultivation implies accumulated knowledge in rice production. Majority of respondents also possess pieces of land for rice farming of more than 1 hectare size and a few had less than 1 ha, and these are youths who are trying to engage themselves in farming activities to increase their sources of income.

Rice yield in the surveyed villages varied significantly. Yields in both villages are still very low compared to the potential yield of (7 t/ha.). Similar results were also reported in Kilimanjaro, Tanzania by Mghase *et al.* (2010). Low yields may be associated with the major constraints of rice production in the respective villages including yellowing and stunting of rice, lack of inputs such as quality seeds, insect pests, diseases, animal pest, and drought. This corresponds to the study by Msangya and Yihuan (2016), which reported that 98% small-scale farmers in Kilombero District in Morogoro region agreed to have challenges in farming while 2% of the farmers said that they did not have. The major challenges which small-scale farmers encountered were high cost of cultivation services during ploughing, unfavourable market, plant diseases, and infestation of weeds, lack of extension services, lack of credit facilities as well as lack of improved varieties.

Significant difference was observed between surveyed villages on type of minor crops produced apart from rice and cropping systems. Mingo farmers practices sole cropping system by producing rice only, while Kimambila farmer's practices mixed cropping of rice as a major crop, and sunflower and maize as minor crops. Sunflower is also used as minor crop at Mingo village by 38.5% of farmers who practiced mixed cropping. Farmers practice mixed cropping in order to obtain extra yield for their home consumptions. Kombiok (2012) suggested that when intercropping and mixed cropping are used in the field it helps to manage insect pest and diseases, maximize production, encourage efficient use of soil resources and diversify crops as compared to mono-cropping.

Among respondents, some reported that rice moderately contribute to their income and few reported that rice contribute less to their income. These farmers largely produce rice for their own consumption and they sell in order to obtain money to acquire other basic needs. Colvin *et al.* (2011) reported that rice was among the top three important crops in terms of income generation to farmers due to its demand for food and as a commodity sold in order to obtain money to acquire other basic needs. Report of URT (2009) shows that majority of rice farmers are smallholders who produce rice for home consumption and sale surplus directly to customer or through a cooperative society where there is a Warehouse Receipt System in operation.

Farmers' rice price is determined through bargaining with the buyers, whereby in most cases buyers set the rice price to small-scale farmers. This situation has made farmers to be economically stagnant as they are not getting benefits from the production of rice. Lack of formal price system, reliable market and poor quality of rice produced add to such low price.

The yellowing and stunting of rice in Kimambila and Mingo villages has only been observed in the past 3 years and it has adversely affected rice production in the villages. The high incidence of the syndrome may be attributed to the lack of knowledge of causal agent(s), lack of control measures for the problem and from farmers' perspective they associated it with poor quality farmers saved seed, susceptible varieties (Supa and Mbawambili), drought and witchcraft. Based on farming experience, the rice farms in Mingo and Kimambila have been used for quite a long time without fallowing or fertilization and they might therefore be nutrient deficient which limit rice production and causing yellowing of leaves and stunting of plants. Pham *et al.* (2007) in a study of etiology of rice yellowing syndrome experienced several symptoms such as serrated, twisted and malformed leaves, vein swelling on leaf sheaths and blades, leaf yellowing, curling and stunted growth and serological test was conducted to identify the cause. Yellowing and stunting of rice are also associated with soil nutrient deficiency as reported by several studies .Rice with nitrogen deficiency usually has no significant symptoms before tillering stage but after the tillering stage it results in stunted and slow growth, and chlorosis with light green to yellow appearance of leaves, especially older leaves (Chen and Wang, 2014).

Respondents also reported on major constraints of rice production in their respective villages and they said the major problems were yellowing and stunting of rice, insect pests, diseases, animal pest, and drought. The most susceptible varieties to yellowing and stunting syndrome as mentioned by respondents in both villages were Supa and Mbawambili in both villages. This concurs with the study by Msangya and Yihuan (2016) who reported that 98% small-scale farmers agreed to have challenges in farming while 2% of the farmers said that they did not have challenges. The major challenges which small-scale farmers encountered were high cost of cultivation services during ploughing, unfavourable market, plant diseases, and infestation of weeds, lack of extension services, lack of credit facilities as well as lack of improved varieties.

Most rice production in the country is rain fed and annual rainfall variations make rain fed rice production susceptible to flooding or drought. The risk of drought impedes investment; causing production to stagnate at a subsistence level (EUCORD, 2012). The lack of availability of rice varieties with tolerance to drought, major insect pests and diseases are major challenges facing the rice sub-sector in Tanzania (URT, 2009). Yield loss, and therefore profit for rice farmers in Kimambila and Mingo villages in Mvomero District is being adversely affected by yellowing and stunting of plants. Based on these observations it seems compelling that the etiology of the condition be elucidated. Only then can measures for management be tested and eventually recommended.

2.6 Conclusion

Generally, lack of information and knowledge on the cause of yellowing syndrome were the main reasons limiting the effective management of the problem. The use of susceptible rice varieties and lack of improved agronomic practices were additional constraints. Our study has provided base-line information on the current status of farmers' knowledge on rice yellowing and stunting of plants in Mvomero district. This information can be used to initiate a process to identify the abiotic and/or biotic causal agent(s) of rice leaf yellowing and plant stunting which will in turn be the basis for developing appropriate management measures for improved rice yield.

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CHAPTER THREE

3.0 IDENTIFICATION AND ANALYSIS OF FACTORS CAUSING LEAF YELLOWING SYNDROME AND LOW YIELD OF ORGANIC RICE (*Oryza sativa* L.) IN MVOMERO DISTRICT

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3.1 Abstract

Rice (*Oryza sativa* L.) is the main source of food and income to smallholder farmers in Mvomero district. This study was undertaken to identify the causal agent(s) of rice yellowing syndrome. Hence rice seed samples (1kg of each variety) were collected from farmers store following ISTA guidelines, five rice plant leaves were collected randomly from symptomatic plants in the field from Mingo and Kimambila villages. The seeds were assessed for physical qualities attributes and identification of seed borne pathogens was done using eight local rice varieties. Blotter method and agar plate method were used to identify seed borne pathogens. A total of six fungal pathogens, (*Bipolaris oryzae, Fusarium moniliforme F. palidiospore, Nigrospora oryzae* and *Phoma* sp), were identified. *Bipolaris oryzae* and *F. moniliforme* were predominant in all tested rice varieties. The seeds with the lowest incidence of seed borne fungi had the lowest incidence of rotten seeds, dead seeds and highest seed germination and seedling vigour index. Among the varieties tested, Supa was the best by having the lowest incidences of seed borne pathogenic fungi, rotten seeds, dead seeds and highest germination percentage and seed vigour. Physical qualities such as purity, germination percentage, seed vigour and 1000 seed weight were also assessed and results showed they align with ISTA recommendation therefore suitable for production. Isolation of fungi and biochemical tests of the pathogens were conducted from randomly selected rice plant leaves and pathogen was detected. Furthermore, 10 composite soil samples were collected using zig zag method from randomly selected farmers' fields in both villages and analysed for physical and chemical properties. Soil pH was found suitable for rice production, Nitrogen and Potassium was very low therefore deficiency. Phosphorus availability rated medium which is sufficient. The main cause of yellowing syndrome after analysis could be due to nitrogen deficiency. Measures needs be taken to increase the availability of macronutrient in the soil to enhance rice production. Uses of organic manure, foliar organic fertilizers together with other sustainable agronomic practice such as fallowing, intercropping, and planting leguminous crops in the field during off season will adjust fertility of the soil.

Key words: Agar plate method, Blotter method, Seed borne pathogens, Soil Nutrients.

3.2 Introduction

Rice (*Oryza sativa* L.) is now only after maize as an important staple food crop for Tanzanian population. Its production is increasing more than any other crop (Agritrade, 2012*)*. It is now grown on more than 700, 000 ha which is about 23% of the land suitable for growing rice. Mbeya, Morogoro,Mwanza, Shinyanga, Coast, , Arusha and Rukwa are the main rice producing regions (Kanyeka, 2001). Yield of the crop in the country is very low (1.5-2t/ha) (Luzi–Kihupi and Zakayo, 2001) while the potential yield can exceed 7t/ha. The low productivity is mostly because of the poor technologies that include use of varieties with low yield potential and poor agronomic practices; and usually without any other material inputs such as soil fertility improvement, weed management and pest and disease control.

Rice is affected by various diseases most of which are seed-borne; fungi and bacteria are pathogens that affect rice at large quantity as compared to viruses and nematodes (Ora *et al.,* 2011). A new rice leaf yellowing syndrome was recently reported by farmers at Mvomero district. The symptoms include but not limited to leaf yellowing, plant stunting, incomplete panicle emergence, plant wilting with subsequent black colouring of the plant and, severe yield reduction. These symptoms closely relate with those of RYMV, Rice orange leaf disease (ROLD), RYDD and Rice yellow stunt disease (RYSD). However, multiple infections can also result into symptoms akin to those of RLYD as reported for co-infection of bacteria and viruses or nutrient deficiency diseases (Luzi-Kihupi *et al.,* 2000; Phuongl *et al.,* 2011).

Soil fertility is also one of the factors affecting rice production as it is diminishing gradually due to soil erosions, loss of nutrient, accumulation of salts and other toxic elements, water logging and un-balanced nutrient compensation. Nitrogen is one of the most limiting rice growth and yield nutrient (Mghase *et al.,* 2010). It is needed by plants for the production of proteins, nucleic acids (DNA and RNA) and chlorophyll.

When nitrogen is deficient old leaves and sometimes all leaves become light green and chlorotic at the tip. Leaves die under severe stress. Except for young leaves, which are greener, N-deficient leaves are narrow, short, erect, and lemon yellowish. The entire field may appear yellowish. Nitrogen deficiency often occurs at critical growth stages such as tillering and panicle initiation, when the demand for N is large (Dobermann and Fairhurst, 2000).

Most rice producers in Kimambila and Mingo are small holder farmers whose access to advanced production technology is limited. These farmers mostly use farm saved seed for raising the next year crop, these seeds are normally of low yielding, impure and contaminated with pathogens. Naher *et al.* (2016) reported that lack of healthy rice seed as one of the most important constraints to rice production and productivity. Seed healthy testing is therefore an important component in the field as health seeds will more likely give out healthy seedlings and thus healthy crops and bumper harvest.

The goal of this study was to identify the main causes of leaf yellowing syndrome and low yield of rice. The study aim to ascertain whether the soil and rice seeds factors had influence on the yellowing syndrome and low yield of rice in Mingo and Kimambila villages.

3.3 MATERIALS AND METHODS

3.3.1 Location of the study area

Laboratory tests to isolate and identify pathogens associated with rice seeds and rice plant and seedling were conducted at the Plant pathology Laboratory at the International Institute of Tropical Agriculture (IITA), Dar-es-Salaam, Tanzania between November-January, 2018. Organic rice seeds, plant leaves and soil samples were collected following ISTA (2003) rules from Kimambila and Mingo villages in Mvomero district, Eastern Tanzania in October, 2018. Mvomero district is located at Latitude: 6° 18′ 0″ South, longitude 38° 41' 37.4928" East and elevation of 400-1500 meters above sea level.

3.3.2 Evaluation of rice seed, plant health and soil fertility

3.3.2.1 Seed sampling and analysis

Eight rice seed samples (1 kg each) were collected from each of eight purposively selected farmers, based on availability of harvested rice seeds in their storage (ISTA, 2003). Seed samples were put in sterile paper bags with correct labelling, brought to the Plant pathology laboratory at the International Institute of Tropical Agriculture (IITA), Dar-es-Salaam and stored at room temperature until testing. The seed samples were analysed for physical characteristics and seed health.

Weight of 1000 seeds was determined using a digital electronic weighing balance with accuracy of 0.001g. To determine the average weight of 1000 seeds, a sample size of 100 seeds were randomly selected and weighed. The result was then multiplied by 10 so as to get the weight of 1000 seeds. Readings were repeated in five times to reduce error. Similar method was reportedly used by Garnayak *et al.* (2008).

Seed purity test was conducted to determine the identity of various species of seeds and inert particles in the samples. The seed sample was separated into three components: pure seeds, other crop seeds (identified by their scientific names), and inert matter. The percentage of each fraction was determined by its weight, calculated to one decimal place. Components of less than 0.05% were reported as trace.

Seed vigour testing was done as follows; four hundred seeds (400) were plated in four replicates of 100 seeds each and incubated in germinator at 24°C for 14 days. The tests were done along with regular germination test. The number of normal seedlings, germinated on the first count of 7 days, as specified in the germination test for rice specie which takes 14 days of growth duration was counted. The seedlings were grown in the laboratory by using rolled paper towels method. Ten seeds were plated in the centre of the moist towel papers with micropyles oriented towards the bottom to avoid twisting of the roots.

The rolled paper towels were kept in the germinator maintained at a temperature recommended for rice which is 24°C. After 14 days of incubation, the paper towels were removed, and five (5) seedlings were selected, their length were measured and mean seedling length was calculated. Seed lots producing taller seedlings were considered vigourous than the seed lots producing shorter seedlings. The seedlings were dried in an air oven at 100°C temperatures for 24 hours. This procedure of dry weight also provides additional information for assessing seed vigour.

Seed vigour index (S.V.I) was calculated by determining the germination percentage and seedling length of the same sample or seed lot as per Kharb *et al.* (1994). Fifty seeds in four replications were kept in rolled paper towels for germination test. Evaluation of number of normal seedlings at the time of final counts was done and seedling lengths of five randomly selected seedlings were also measured, and their mean recorded.

Seed vigour index was calculated following Equation 2:

$$S.V.I = Germination\ percentage(\ )\ X\ Seedling\ length(mm) \quad \ldots\ldots\ldots\ldots (2)$$

Therefore, the seed lot or seedling showing the higher seed vigour index was considered to be more vigourous (Abdul-Baki and Anderson, 1973).

Seed moisture content was determined by subjecting seeds to oven drying and measuring the weight of samples at temperatures of 130ºC for 8hrs to 24hrs by ASAE standards (ISTA, 2003). The dry weight of seeds was calculated using the formula summarized in Equation 3.

$$Dry\,weight\,of\,seeds = \frac{Weight\,before\,drying - Weight\,after\,drying}{Weight\,after\,drying} \quad \ldots\ldots\ldots\ldots\ldots\ldots$$

(3)

One hundred seeds in four replications were planted in rolled paper towels for germination test. The paper towels were placed in vinyl plastic bags to avoid moisture evaporation during germination, and kept in the germinator maintained at recommended temperature for rice which is 24°C. Number of seedlings emerging daily were counted from day of planting the seeds in the medium until the duration of germination was completed, which is 14 days for rice. Then, a germination index (G.I) was computed by using a formula indicated in Equation 4:

$$G.I = \frac{n}{d} \quad \text{x } 100 \ \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots$$

(4)

Where n = number of seedling emerging on a day “d”

d= days after planting.

Identification of seed borne pathogens was done by using Standard Blotter Method and Agar Plate Methods, following the rules for seed testing of the International Seed Testing Association (ISTA, 2003). A total of 200 seeds were randomly picked from each of the eight rice seed samples.

Before conducting testing, the seeds were surface sterilized by soaking in 10% sodium hypochlorite ($NaOCl_2$) for one minute followed by rinsing three times with sterile distilled water. Two hundred seeds were taken from each of the eight samples and plated on moistened blotter at 25 seeds per Petri dish in a pattern of 16 seeds at the outer ring, eight seeds in the middle and one in the center. The cover of each Petri dish was put back and securely held in place with a Parafilm. Then the Petri dishes were incubated in a growth chamber at temperature of 22^{o}C for 7 days at alternating cycle of 12 hours of darkness and 12 hours of light (ISTA, 1998). After incubation, fungi that developed on each seed were examined under different magnifications of a stereomicroscope and identified. Growth habit and morphological characters of fruiting bodies, spores/conidia as observed under a compound microscope were used as identification criteria (Mathur and Kongsdal, 2003).

In agar plate method, 200 seeds were surface sterilized by soaking in 10% sodium hypochlorite ($NaOCl_2$) for one minute followed by rinsing three times with sterile distilled water. Seeds were plated on Potato Dextrose Agar (PDA) for observation of associated pathogens. Twenty five (25) seeds were plated per Petri dish and eight Petri dishes were used per sample. The seeds were arranged on PDA in 9cm Petri dish. The Petri dishes were incubated in a growth chamber at 22^{o}C for seven days at alternating cycles of 12 hrs light and 12 hrs darkness. After the incubation period, fungi that developed from the seeds on the Agar medium were examined under stereomicroscope and identified based on colony characteristics and morphology of spores and fruiting bodies under different magnification of a compound microscope and by comparing with available literature (Mathur and Kongsdal, 2003).

3.3.3 Plant sampling and seedling analysis

3.3.3.1 Plant analysis

Leaves were randomly collected from the symptomatic and asymptomatic plants and brought to the Plant Pathology Laboratory at the Crop Science and Horticulture Department, SUA, Morogoro. Collection of the leaves involved a number of infected and health leaves from each of the eight samples.

Rice leaves from the field experiment with yellow color and other discolorations were selected for isolation from five rice plants exhibiting the yellowing of leaves. Infected leaves were surface sterilized by dipping them into 95% ethanol for 1 minute and then treated with NaOCl 1% solution for 2 minutes and rinsed 3 times with sterile water to remove residual hypochlorite.The infected pieces with symptoms of bacterial infection were cut and placed onto a glass slide in a drop of water.

A compound microscope was used to gaze for ooze from bacteria. Pieces showing bacterial ooze were cut into smaller pieces in a drop of saline and left for 15 minutes so as to obtain more bacterial cells in the water. Using a bacterial loop the water containing bacterial cells was streaked onto 3 plates in replicate containing Nutrient Agar (NA) in order to get single separate colonies (Fig. 3.1).

Figure 3.1: Bacterial ooze streaming from excised plant tissues

The streaked plates were then incubated in an inverted position at 25^0 C. Plates were observed after 2 days for *Pseudomonads* and up to 4 days for *Xanthomonas*. Suspected yellow colonies in (Fig. 3.2) were then transferred to another plate for purification.

Single colonies were selected and streaked onto other plates by flaming the loop in between streak in order to obtain separate colonies for identification as shown in (Fig. 3.3). Single colonies had to undergo biochemical tests for identification of bacteria.

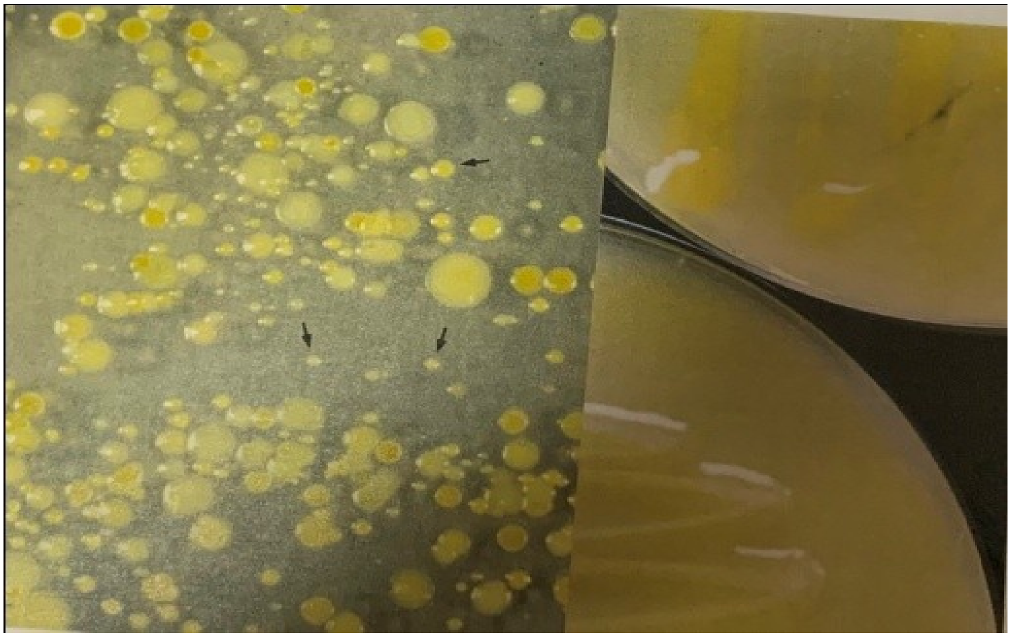

Figure 3.2: **Suspected colonies of *Xanthomonas oryzae pv. Oryzae***

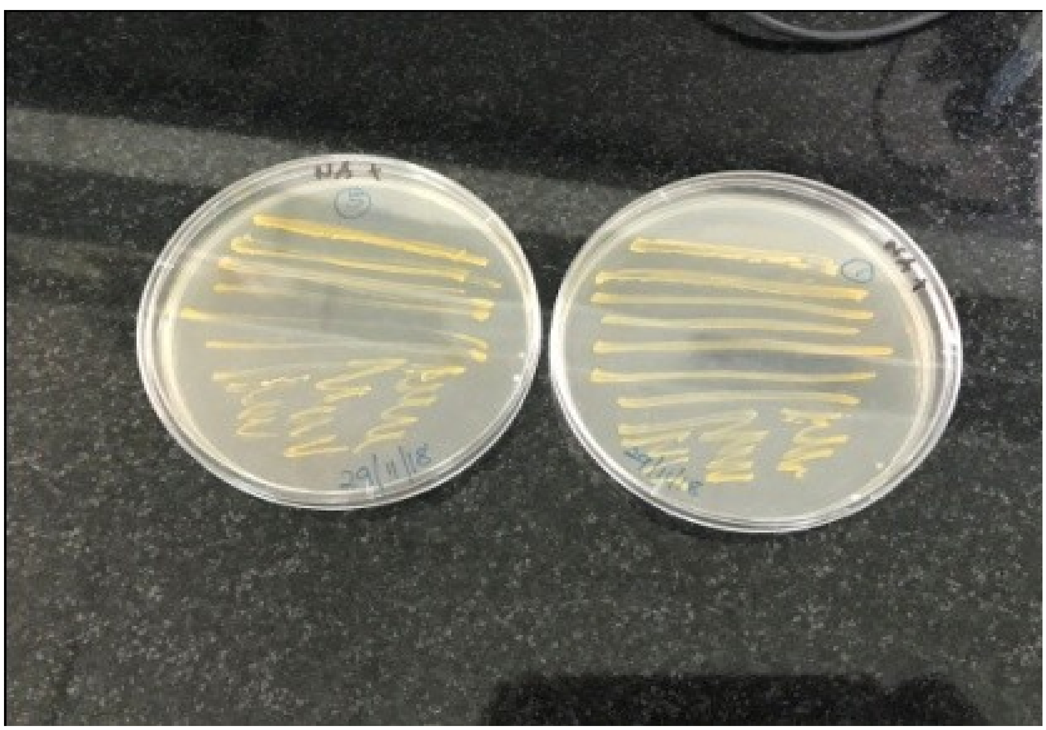

Figure 3.3: **Plates for samples # 1 and # 5 showing yellow colonies**

3.3.4 Biochemical tests of the pathogen

3.3.4.1 Potassium hydroxide solubility test

On a glass slide a loopful of bacteria from a well grown colony was mixed in a drop of KOH aqueous solution. Mixing didn't exceed 10 seconds. A loop was used for picking bacteria from a colony as well as mixing, where a toothpick could also be used. A loop was raised a few centimetres from the glass slide. Strands of viscid material were observed.

3.3.4.2 Kovac's oxidase test

A Whatman filter paper No. 1 was placed in a Petri dish and added 3 drops of freshly prepared 1% aqueous solution of N- N-dimethyl-p-phenylenediamine on the centre of the filter paper. With a help of a platinum loop, a loopful of bacteria, grown on NA was streaked on the moist filter paper to observe colour change of the reagent.

3.3.4.3 Starch hydrolysis

Starch agar media plates were inoculated by streaking the bacterium colony and incubated for 4 days**,** flooded with Lugol's iodine**.** Positive reaction was indicated by an appearance of yellowish, clear zones around or under the bacterial growth an indication of amylase activity.

3.3.4.4 Gelatin hydrolysis

Test tubes containing gelatine medium were stab-inoculated including an un-inoculated as a positive control. The test tubes were incubated at 20°C for 7 days.

Every 2-3 days record of liquefaction of the medium was monitored. The test cultures were left to solidify. The cultures were then removed and the negative control incubated in a tilted position at room temperature for about 30 minutes.

3.3.4.5 Arginine dihydrolase test

Inoculation was done by stabbing with a fresh culture tube containing 3 ml of Thornley's medium**,** covered with mineral oil and incubated for 3 days at 27°C. A change of colour in red (alkaline) is a positive reaction and no change indicates negative reaction

3.3.5 Soil sampling and analysis

The method by Thiagalingam (2000) was used, whereby 10 composite soil samples were collected in each farm from 10 representatives among interviewed farmers. Each of the 10 composite samples was made up of six sub samples collected on a Zigzag sampling configuration and at the depth of 0 – 20 cm in each farm. The composite soil samples were reduced by the quartering procedure to obtain a representative sample weighing 0.5kg and then passed through a 2mm aperture sieve for laboratory analysis.

The soil samples were analyzed for physical and chemical properties at the Soil and Geological Science Laboratory at Sokoine University of Agriculture (SUA), Morogoro. The samples were air-dried, ground with a wooden pestle and mortar and passed through a 2 mm sieve. Soil pH was measured in water using a pH meter at the ratio of 1:2.5 soil: water as described by McLean (1982). The CEC was determined using Ammonium Acetate saturation method as explained by Chapman (1965). The particle size analysis was determined by the hydrometer method after dispersion with sodium hexametaphosphate as explained by Day (1965). The textural class was determined using the USDA textural class triangle (USDA, 1975).

Total Nitrogen was determined Micro-Kjeldahl digestion-distillation method as explained by Bremner and Mulvaney (1982). Available P was extracted by the Bray and Kurtz-1 procedure (Bray and Kurtz, 1945), for soil soils with pH water less than 7 and Olsen methods for soils with pH water above 7 and determined spectrophotometrically following colour development by the molybdenum blue method (Murphy and Riley, 1962; Watanable and Olsen, 1965).

DTPA extractable micronutrients in all soil samples were determine using procedure described by Lindsay and Norvell (1978). The extractant contained 0.005M DPTA (Diethylenetriaminepentaacetic acid), 0.01M $CaCl_2$, H20 and 0.1M TEA (Triethanolamine) adjusted to pH 7.3. Fifteen grams of air dried soil were mixed with 20ml of extracting solution and shaken for two hours and then filtered through Whatman No. 42 filter paper. Concentration of Zn, Cu, Fe and Mn in the filtrate was determined with atomic absorption spectrophotometer.

Soil organic carbon was established by wet digestion method of Walkey and Black (1934) cited by Nelson and Sommers (1982). The CEC was determined by the buffered ammonium acetate saturation method (Chapman, 1965). The quantity of exchangeable bases K+ and Na+ was determined by Flame photometer while Ca2+ and Mg2+ were assayed by Atomic Absorption Spectrophotometer (AAS) as described by Thomas (1982).

3.4 Results and discussion

3.4.1 Physical characteristics and mycoflora in rice seeds

Table 3.1: Physical characteristics of rice seed samples from Mvomero district

Sample number	Seed purity (%)	Seed vigour index	Germination (%)	Moisture content (%)	1000 seed weight (g)
1	98.3	745.5	82	7.40	27.628
2	99.2	706.8	92	7.41	23.985
3	95.1	564.4	92.7	7.60	28.940
4	97.3	446.6	82	8.15	31.436
5	97.9	940.5	85	8.91	27.453
6	99.2	819.0	85.5	8.07	28.976
7	99.3	806.4	85.7	7.36	32.549
8	99.1	592.8	79.2	7.58	29.786
Mean	98.18	702.75	85.51	7.81	28.84
Maximum	99.30	940.50	92.70	8.91	32.55
Minimum	95.10	446.60	79.20	7.36	23.99
SD	1.44	160.12	4.76	0.54	2.63

The results (Table 3.1) revealed that farmers saved rice seeds possessed quality physical characteristics suitable for rice production. Seed purity percentage was high for all samples and ranged from 99.3% to 95.1%. These values are in line with the ISTA (2003) recommendations which suggested that the percentage purity for rice should be at least 96%. Gebeyehu *et al.* (2019) also reported 80 percent of rice seed samples from farmers to have more than 90% purity. Poor quality of rice seeds often results from infection by pathogens, weeds, insect pests and rice admixtures. Using poor quality rice seeds for planting reduces the productivity of modern cultivars in attaining its genetic potential (Haque *et al.*, 2012).

Seed vigour index for rice samples ranged from 446.6 (which was the lowest) to 940.5 the highest. Seed vigour is one of the key components of seed quality that provides accurate information on the field performance potential of seed lots. High vigour seeds can resist the negative impact of variable environmental conditions during production and processing. Crop yield and resource use efficiency depend on successful plant establishment in the field, and it is the vigour of seeds that defines their ability to germinate and establish seedlings rapidly, uniformly, and robustly across diverse environmental conditions. Maturity stage in harvesting time greatly affects seed vigour (Wang *et al.,* 2018). A combination of higher crop emergence, vigorous early crop growth, and increased crop resistance to insect pests and diseases will result to a 20% increase in yield (IRRI, 2002). Reports shows that poor quality seed gives poor seedling vigour, non-uniform growth and maturity and further prone rice seedlings to insects and disease and weed-pests, and adverse environmental conditions (Dogbe *et al.*,2015).

Germination percentage of rice samples ranged from 79.2 (the lowest) to 92.7 as the highest. Germination percent affects yield and determine the planting value of seed. Talei *et al.* (2013) reported that average germination percent ranged from 93% to 98% in different rice varieties exposed to different temperature The level of germination in association with seed vigour provides a very good estimate of the potential field performance (Gummert, 2010). When seed is stored in traditional open systems, the germination rate of most rice seed begins to deteriorate rapidly after six months. Also, many varieties have a dormancy period immediately after harvest that can last for 1−2 months (Gummert, 2010). The propagation of rice generally occurs through seeds or seedlings, and seed germination depends on seed structures and environmental factors that affect the growth potential of embryo (Koornneef *et al.,* 2002). By knowing the germination rate, farmers can adjust their planting rates to attain the desired plant population in the field

Seed moisture content values ranged from 7.36 % to 8.91% among tested rice samples. Moisture in seeds is an important part of seed testing as it helps to determine the optimum time for harvest and identifies appropriate seed storage and drying recommendations. A study by Alhendi *et al*. (2019) showed that moisture content of rice in two tested varieties 14% MC and 10% MC of Yasemin and Anber respectively were the best MC in terms of producing high extraction rate, lower kernel breakage, and high rice elongation. The initial moisture content of paddy had an effect on the cooking properties of the produced rice

Weight of 1000 seeds ranged from 27.45 to 32.54 g in tested rice samples. According to ISTA the average 1000 seed weight of paddy is 19.18g. There are various factors that can affect 1000 seed weight such as genetic features, nominal potential or sprouting potential, seed potential, humidity, storage quality, seed preservation and health, but the most important ones are sprouting rates and seed potential (Malcolm *et al.,* 2003; Akbari *et al.,* 2004). Test weight is an important factor to consider when selecting a variety as both environment and pests may greatly affect test weight therefore, selecting a variety that has a high test weight potential in any area is critical to maximizing economic gain (Deivasigamani and Swaminathan, 2018). Further studies also reported that, 1000 seed weight has significant effects on sprouting percentage and dry seeding weight, while it has no significant effect on seedling length (Mashtati *et al.,* 2008).

The results (Fig. 3.4) show that, various fungi were detected after seed pathogen test. A total of five seed borne pathogens were identified. These were *Fusarium moniliforme, Bipolaris oryzae, Nigrospora oryzae, Phoma spp and F. palidoroseum.* Among the observed pathogen *Fusarium moniliforme,* had the highest frequency of occurrence in all samples (48.89%) which is about half of the samples and *F. palidoroseum* had the lowest frequency (0.80%) as it occurred in three samples only but had the highest frequency as compared to other fungi.

On the other hand using agar plate method, only *F.moniliforme* and *F. palidoroseum* were observed in the samples. *Fusarium moniliforme* had the highest frequency 97.9% in all samples and *F. palidoroseum* occurs by 2.07% only. Of all the fungal pathogens detected *F. moniliforme* and *F. palidoroseum* were predominant in all samples. Ora *et al.* (2011) reported that *Xanthomonas spp, Rhizopus stolonfers, Bipolaris oryzae and F.moniliforme* were predominant from their experiment on seed mycoflora of rice. Rahman *et al.* (2010) using agar plate method identified major genera of fungi detected from the experiment were *Bipolaris, Fusarium, Curvularia, Alternaria* and *Aspergillus.*

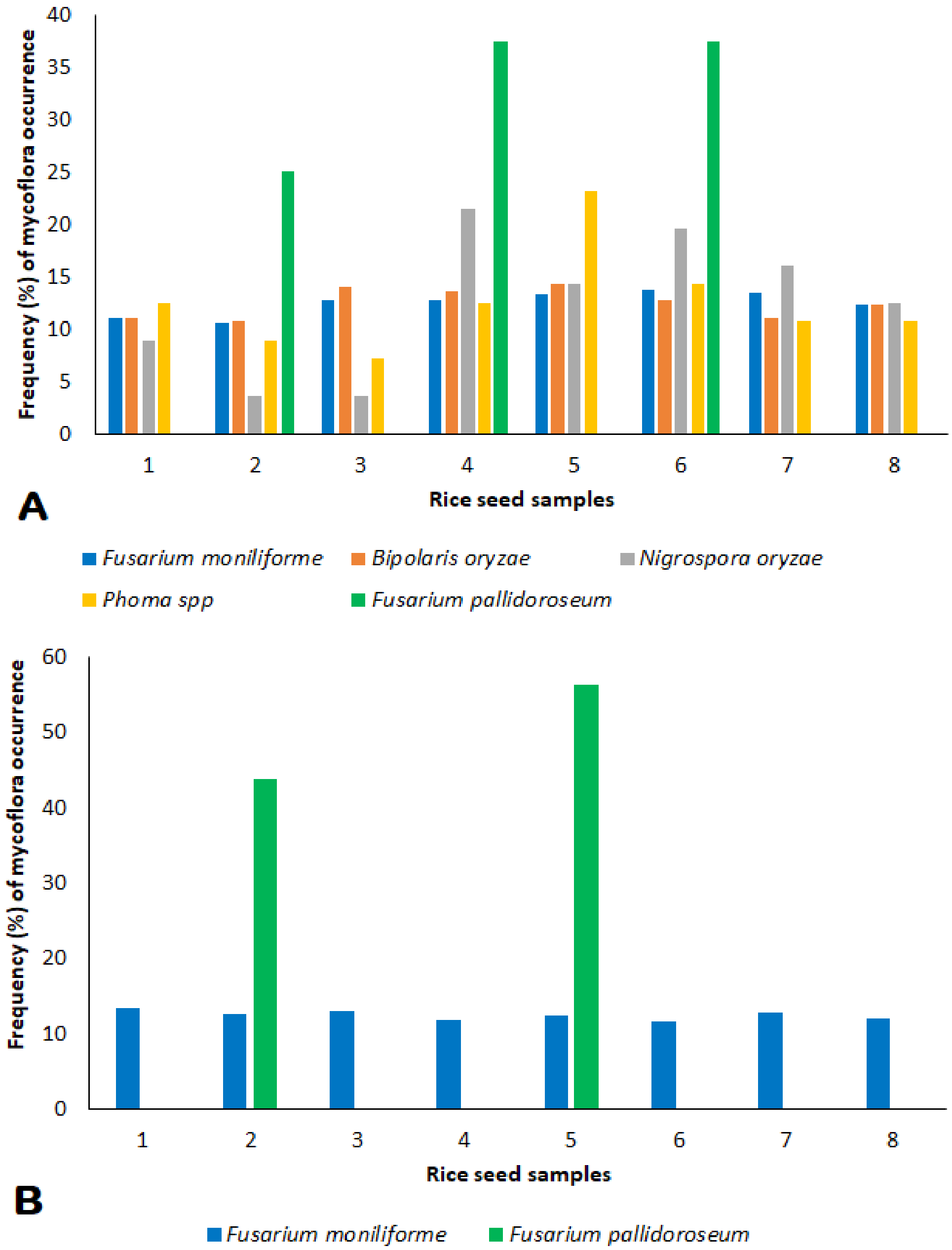

Figure 3.4: **Frequency (%) of occurrence of seed borne mycoflora in different organic rice seed samples.** A. **Detected by blotter paper method.** B. **Detected by Agar plate method.**

Bipolaris oryzae is the fungal pathogen causing brown spot disease of rice, it is observed in infected seeds and plant parts. Frequency of detection is between 1-65% in most seed samples and it has low epidemic potential (IRRI, 2002). Reports show that severe infection can reduce yield by 20-40% and abnormal soil conditions increase damage. Seed treatment and proper agronomic management practices may help to manage brown spot disease. These results reveal that seed borne pathogens are present in most of cultivated varieties and are probably the major cause of diseases in the field. Under suitable conditions and susceptible host these pathogens may cause disease in epidemic level. Using pathogen-free seeds is vital as the primary input in production.

Storage conditions and life cycle of pathogens may have favoured the occurrence of pathogens in the seeds. Various methods may be used by farmers to reduce incidence of disease occurrence example by treating the seeds with fungicides, and improving storage conditions. IRRI (2002) narrated physical and chemical methods for seed treatments. Upon dry inspection of seeds some other aspects were also observed such as weed and dead insects. Rice seeds contaminated with weed seeds may introduce new species or add on existing weed seed bank in the field. *Fusarium moniliforme* is the causative agent of bakane or foot rot disease in rice. Loss in yields due to the disease ranges between 3.7-50%.

4.4.2 Relationship between the physical properties of rice seeds and frequency of mycoflora occurrence as detected by blotter paper method

The results (Table 3.2) indicate a significant (R^2 = 0.98, p = 0.04) relationship between the frequency (%) of mycoflora (*Phoma spp*) occurrence as detected by blotter paper method and the seed physical properties namely; seed purity (r = 0.16), moisture content (r = 0.88), seed vigour index (r=0.64), germination (r = -0.35) and 1000 seed weight (r = -0.07). However, there was no significant relationship between the physical properties and frequency of *Fusarium moniliforme* (R^2 = 0.90, p = 0.23), *Bipolaris oryzae* (R^2 = 0.93, p = 0.17), *Nigrospora oryzae* (R^2 = 0.90, p = 0.23) and *F. pallidoroseum* (R^2 = 0.61, p = 0.7) occurrence as detected by blotter paper method.

Table 3.2: Correlation (r) between the physical properties of rice seeds and frequency (%) of mycoflora occurrence as detected by blotter paper method

	Fusarium moniliforme	***Bipolaris oryzae***	***Nigrospora oryzae***	***Phoma spp***	***F. pallidoroseum***
Seed purity (%)	-0.08	-0.67	0.31	0.16	0.18
Seed vigour index	0.25	-0.14	0.05	0.64	-0.26
Germination (%)	-0.14	0.03	-0.63	-0.35	0.05
Moisture content (%)	0.51	0.76	0.48	0.88	0.19
1000 seed weight (g)	0.67	0.19	0.66	-0.07	-0.06
Regression (R^2)	0.90*	0.93*	0.90*	0.98**	0.61*
p-value	0.23	0.17	0.23	0.04	0.70

** = Significant (p<0.05) relationship, * = Relationship not significant (p>0.05).

4.4.3 Relationship between the physical properties of rice seeds and frequency of mycoflora occurrence as detected by Agar plate method

There was no significant ($p > 0.05$) relationship between frequency (%) of mycoflora (*Fusarium moniliforme* and *F. pallidoroseum*) occurrence as detected by agar plate method and the physical properties namely; seed purity, moisture content, seed vigour index, germination, 1000 seed weight (Table 3.3).

Table 3.3: Correlation (r) between the physical properties of rice seeds and frequency (%) of mycoflora occurrence as detected by Agar plate method

	Seed purity (%)	**Seed vigour index**	**Germination (%)**	**Moisture content (%)**	**1000 seed weight (g)**
Fusarium moniliforme					
Correlation (r)	0.26*	0.15*	0.34*	0.48*	0.25*
Regression (R^2)	0.07	0.02	0.12	0.23	0.06
Intercept	22.72	12.13	8.87	16.64	-0.06
Slope	-0.10	0.00	0.04	-0.53	14.14
Std. Error	0.16	0.00	0.05	0.39	0.088
p-value	0.54	0.73	0.41	0.22	0.54
F. pallidoroseum					
Correlation (r)	0.12*	0.52*	0.33*	0.5*	0.68*
Regression (R^2)	0.02	0.27	0.11	0.25	0.46
Intercept	-185.91	-40.56	-125.02	-159.00	185.89
Slope	2.02	0.08	1.61	21.96	-6.01
Std. Error	6.57	0.05	1.90	15.34	2.68
p-value	0.77	0.19	0.43	0.20	0.07

* = Relationship not significant (p>0.05).

3.4.4 Pathogenic species isolated from rice plant tissues

3.4.5 Biochemical tests of the pathogen

3.4.5.1 Potassium hydroxide solubility test

Strands of viscid material were seen as shown in Figure 3.4 the bacterium in question is gram-negative, where gram-positive bacteria do not produce such strands.

3.4.5.2 Kovac's oxidase test

With a help of a platinum loop, a loopful of bacteria, grown on NA was streaked on the moist filter paper. Unchanged colour of the reagent didn't take place within 10 seconds of application of the culture indicating negative test result, as shown in Fig. 3.5 whereby the bacterium in question was negative to the test (Kovac's, 1956; Hildebrand and Scroth, 1972)

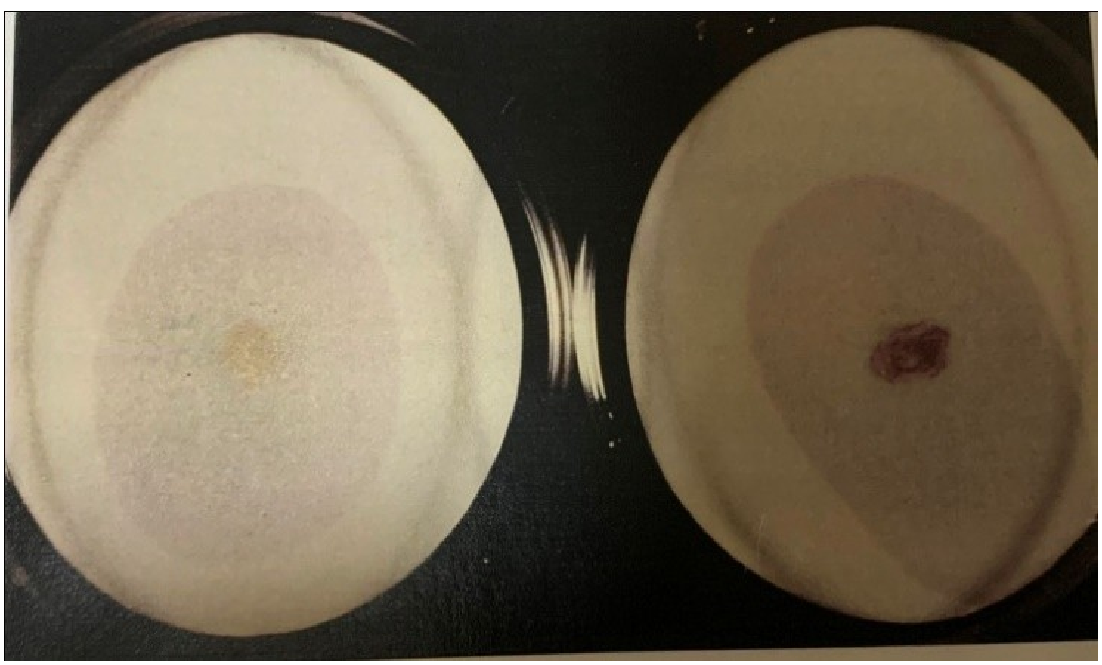

Figure 3.5: **Oxidase test: Left negative; Right, positive**

3.4.5.3 Starch hydrolysis

This is the ability of certain bacteria to hydrolyse starch. It is a method used to differentiate bacteria in a taxonomic character. Positive reaction was indicated by an appearance of yellowish, clear zones around or under the bacterial growth an indication of amylase activity. Starch staining blue in a medium, starch has not been hydrolysed, negative reaction as our bacterium in question has indicated as shown in (Fig. 3.6) left.

Figure 3.6: **Starch hydrolysis. Left, negative; Right, positive**

3.4.5.4 Gelatin hydrolysis

Proteolytic bacteria decompose gelatine with loss of gelling properties. This test is shown as a test in which the organism grows in a nutrient medium with solidified gelatine. The inoculated test culture took as long as the un-inoculated control for liquefaction as shown in (Fig. 3.7**)** indicating negative reaction as to the bacterium in question has indicated (Fahy and Persley, 1983: Lelliot and Stead, 1987).

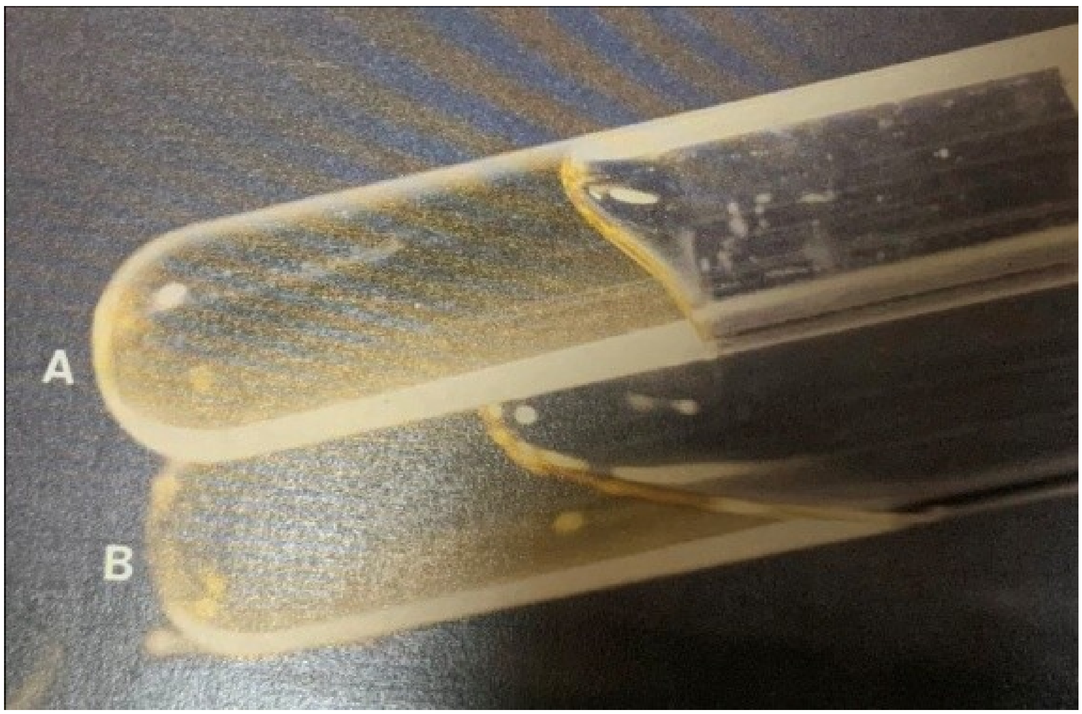

Figure 3.7: Gelatin hydrolysis. A: negative reaction; B: positive reaction

3.4.5.5 Arginine dihydrolase test

The arginine dihydrolase system permits certain bacteria to grow under anaerobic conditions. Ammonia is evolved which brings about the change in pH, indicating a positive reaction, and if no changes indicating negative as shown in (Fig. 3.8) right. A change of colour in red (alkaline) is a positive reaction and no change indicates negative reaction as our bacterium in question has indicated.

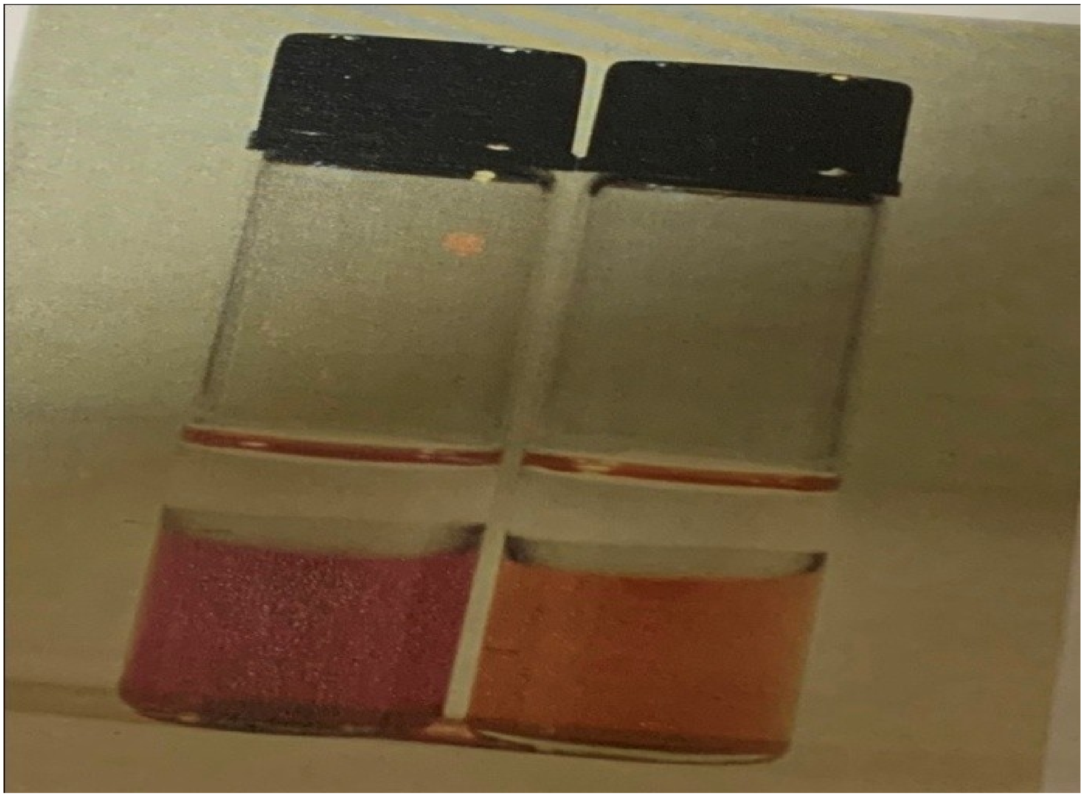

Figure 3.8: Arginine dihydrolase test: left, colour change to red (positive), right, unchanged colour, (negative).

3.4.6 Status of soil nutrients in rice farms

3.4.7 Physical and chemical characteristics

3.4.7.1 Soil texture, pH and Organic carbon content

Physical and chemical properties of soil samples collected from rice fields in the study area are summarized in Table 3.5.

The soil textural class was sandy clay loam for Kimambila site with particle size distribution of 23.8%, 2.64% and 73.52% for clay, silt and sand respectively. Mingo site has clays soil textural class with particle size distribution of 65%, 12.46% and 22.52%

for clay, silt and sand respectively. The organic carbon content in the soils averaged 2.43 % at both sites Kimambila and Mingo. The value rated as being low Landon (1991) for rice production as it ranges from 2-4 %. For improvement of the soil condition for upland rice production amendment of the soils should be done through incorporating manure and plant residues in the soils.

Table 3.4: Soil physical and chemical characteristics of the study areas

Parameter	Optimal range	Kimambila	Mingo	Remarks
Nitrogen (%)	0.21-0.50	0.12	0.09	Low
Organic carbon (%)	4-10	2.4	2.4	Low
P(mg/kg)	5-10	2.446	1.863	Low
Clay (%)		23.8	6.5	
Silt (%)		2.64	12.46	
Sand (%)		73.52	22.52	
CEC ($Cmol_{(+)}$/kg)	20-60	17.48	29.8	Medium/high
Boron (mg/kg)	1.5-3	0.91	9.04	Toxic
Ca^{2+} ($Cmol_{(+)}$/kg)	5.1-10.0	9.864	7.476	Medium
Mg^{2+} ($Cmol_{(+)}$/kg)	0.75-2.0	1.92	5.66	Very high
Na^{2+} ($Cmol_{(+)}$/kg)	0.31-0.70	0.8	1.71	Medium
K ($Cmol_{(+)}$/kg)	0.26-0.80	0.60	0.72	Medium
Cu(mg/kg)	1.58-2.98	1.9	3.0	Sufficiency/high
Zn (mg/kg)	0.59-1.75	1.00	1.15	High
Fe (mg/kg)	30-293.42	105.95	148.75	Sufficiency/high
Mn (mg/kg)	15.95-58.06	39.9	28.2	Sufficiency/high
pH (water)	6.6-7.3	6.87	6.66	Neutral
Sulfur(mg/kg)		9.55	9.88	Deficiency

Optimal range (Landon, 2004); Remark (Msanya *et al* ., 2001)

Soil pH of the analysed samples ranged from 6.28 to 7.29 for Kimambila site and thus Neutral. At Mingo it ranged from 6.32-7.02 and therefore Neutral in water as laid down by Landon (1991). Cation exchange capacity (CEC) rated medium at Kimambila 17.48 $Cmol_{(+)}$/kg and high at Mingo village 29.8 $Cmol_{(+)}/kg^{-1}$ as recommended by Msanya *et al. (*2001). According to Landon (2004) both sites had low organic carbon for analysed soil samples with an average of 2.4%.

According to Landon (1991) organic carbon rates <2; Very low, 2-4 Low, 4-10 Medium, 10-20 High and >20 Very high. Organic matter plays a significant role in enhancing availability of other nutrients in the soil. Upon decomposition of organic matter it also helps in availability of micronutrients in the soil. Sulphate –sulphur was deficiency at both sites; values averaged 9.55 for Kimambila and 9.88 for Mingo. Calcium can cause sulphate precipitation and thus reducing S availability. Sulphur deficiency is not common in rice also sulphur fertilization.

3.4.7.2 Macro (N, P, K, Ca+, Mg, SO_4^{2-}-S) nutrients

The results of soil analysis showed that Kimambila and Mingo sites contained very low amount of nitrogen, 0.12 % and 0.09%, respectively. Total nitrogen of soil is usually classified as very high if (>1.0 %), high (0.5 – 1.0 %), medium (0.2 – 0.5 %), low (0.1 – 0.5 %) and very low (<0.1 %) (Dierolf *et al.*, 2001; Landon, 1991). According to the standards suggested by Landon (1991) for soil nitrogen, about 100% of the soil samples were deficient in total N, in all samples for normal rice plant growth. This deficiency might be due to difference in natural fertility of the rice fields' soils, cultural practices such as using the land for long time without using fertilizer, removing and feeding cattle rice straws in the field. Nitrogen deficiency in the soil samples may also be due to the chemical reactions occurring under reduced conditions, i.e. denitrification, ammonia volatilization, clay fixation, losses in run-off water and deep percolation (Nazir *et al.*, 2003). Based on this classification soils from the two sites need addition of nitrogen from either organic or inorganic sources. Nitrogen plays vital role in rice by enhancing formation of leaves which are significant in photosynthesis. Nitrogen in the soil can be added by using inorganic fertilizers incorporated with compost or manure (farmyard or poultry manure) at a recommended rate. Cultural practices such as planting leguminous (e.g. common beans and soya beans) plants and fallowing may be used to improve soil structure and fertility status of the soil.

The available P rated deficiency 2.446 mg kg^{-1} for Kimambila and high 18.63 mg kg^{-1} in Mingo by using Bray and Kurtz-I methods, while by Olsen method was rated medium 22.93 mg kg^{-1} for Kimambila and 9.46 mg kg^{-1} medium for Mingo respectively as shown in the Table 3.4 . Phosphorus is an important nutrient element in plants supporting root growth development. Most crops demand more phosphorous for optimal yields. Therefore, although it is available there is a need of supplying more phosphate in the soil though fertilizer sources including such as DAP, NPK, NAFAKA PLUS (from Minjingu Phosphate mines company Ltd., Arusha, Tanzania) but also incorporated with farmyard manure or poultry at adequate rate.

The exchangeable potassium (K^+) rated low at Kimambila and Medium at Mingo (0.60 and 0.72 $Cmol_{(+)}$/kg) respectively as proposed by Landon (2004). Potassium is very important for grain quality, being deficiency it affects the quality of rice grains at Kimambila and as nitrogen is also deficiency the two may have contribute to rice stunting, yellowing and low yield at Kimambila and Mingo. Adequate availability of potassium in the soil enhances the uptake of magnesium by plants.

Magnesium (Mg^{2+}) at both Kimambila and Mingo was very high 1.92 Cmol $_{(+)}$/kg and 5.66 Cmol $_{(+)}$/kg respectively. For calcium it was rated very high at Kimambila and medium at Mingo 9.864 Cmol $_{(+)}$/kg and 7.476 Cmol $_{(+)}$/kg according to specific soil texture as proposed by Msanya *et al. (*2001). Exchangeable base sodium was very low at Kimambila sites averaged 0.8 Cmol $_{(+)}$/kg and low at Mingo 1.71 Cmol $_{(+)}$/kg. The sulphate-sulphur level of sample soils indicated that rice fields were the same in soil S contents as they were both deficiency as per Landon (2004) standards. Boron result rated toxic for both sites 8.91 and 9.04 mg kg^{-1} for Kimambila and Mingo respectively. Presence of Boron in the soil is associated with evaporation of ground water, therefore always found in high concentration in arid soils. Toxic level is common in saline –sodic soils and occurs mostly in volcanically active areas (Landon, 1991). For soluble Ca+ and Mg they both rated high at Kimambila and medium for Ca+ at Mingo while high for Mg. This might have been associated by the availability of magnesium in the soil. Normally Ca^{2+} availability in the soil is attributed by high CEC, pH >5.5, availability of moisture.

3.4.7.3 Micro (Zn, Cu, Fe, Mn) nutrients

Average contents of DTPA extractable Zn in soil was high for both sites Kimambila and Mingo according to Tisdale *et al.*(1993) cited by Tisdele *et al.* (2014). The DTPA extractable Cu in the soils samples of Kimambila and Mingo were sufficiency 1.9 and 3.0mg kg^{-1} respectively, according to Tisdele *et al. (*2014). Results also indicated that there was no iron deficiency in the sample soils according to the standards outlined by Lindsay and Norvell (1978); all samples had sufficiency /high amount 105.95 and 148.75 mg kg^{-1} for Kimambila and Mingo respectively. Iron deficiencies are not common in flooded rice, because flooding enhanced the reduction and solubility of soil iron (Patrick *et al*., 1985).

DTPA extractable Manganese contents of soils ranged sufficient in both sites Kimambila and Mingo 39.9 and 28.2 mg kg^{-1} respectively. Results indicated that all samples were high and have sufficiency manganese for normal plant growth according to the standards reported by Lindsay and Norvell (1978). This higher amount of Mn in the soils might be due to its higher solubility caused by reduction of Mn following flooding. Micronutrients (Zn, Cu, Fe, and Mn) were all present in sufficient amount at both sites. The high levels of Fe in all soils could be attributed to the low pH and nature of the parent material from which the soil were formed (Alloway and Ayres, 1997). The high levels of Mn in the soils might be due to low pH that favours the dissolution of Mn minerals in soils (Alloway, 1990).

3.5 Conclusions and recommendations

The major cause of yellowing syndrome from these results is low amount of nitrogen in soils of Mingo and Kimambila villages. This deficiency is associated with continuous uses of fields without any nutrient management practices, large groups of animals' movement which deteriorates soil structure, poor agronomic practices and boron toxicity. The current study has also revealed the presence of several seed borne pathogenic fungi associated with rice seed samples from Mingo and Kimambila villages in Mvomero district .The revealed pathogens although are not associated with the yellowing syndrome but indicate the possibility of disease episode when such infected seeds are used to raise the next rice crop. In as much as the results of this study may be considered preliminary, they nevertheless imply fungi associated with rice seeds are potential threat to its production. A further study into the importance of these seed borne pathogens with respect to rice growth and yield is warranted for effective and sustainable management of seed heath and seed quality of rice.

Generally, soil pH suitable for rice production should range from 5.5 -7.0 and samples of soil from Kimambila and Mingo show that the soil is neutral and suitable for rice production. Phosphorus availability rated medium which is sufficient for short term production and after a time it will be used hence the need to practice fallowing and leaving rice straws in the field after harvesting. Potassium was low for Kimambila but also Mingo although it rated medium. As for nitrogen and phosphorus, measures need to be taken so as to increase the availability in the soil to enhance rice production. A study to explore the type and amount of organic fertilizer to be used to retain fertility and other agronomic packages should be conducted. Awareness of the quality of seeds recommended for usage is also profound to farmers so as to reduce risk of contamination and disease occurrence. Resistant varieties with good agronomic characteristics should also be emphasized to be used by farmers at Mvomero district.

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CHAPTER FOUR

4.0 SUSTAINABLE MANAGEMENT STRATEGIES FOR "LEAF YELLOWING SYNDROME" OF RICE (*Oryza sativa* L.) IN MVOMERO DISTRICT

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4.1 Abstract

A study was conducted at Mingo village in Mvomero district, Morogoro region, Tanzania from May 2019 to September 2019, aiming at improving rice performance through enhanced soil fertility using organic fertilizers. A split–split plot experiment in a randomized complete block design with four replications and16 treatment combinations was used. The main factor was four rice varieties namely (i) Mbawambili (ii) Mwangaza (iii) Supa and (iv) Saro. The sub factors was different fertilizer types, (i) Cow dung manure at the rate of 5t ha^{-1}, (ii) Urea 46% N at a rate of 80 kg N ha^{-1} and (iii) Compost at the rate of 5t ha^{-1} and (iv) No fertilizer (control). Rice was sown in 2m x 2m plots each with five rice rows at 20cm inter–and intra–row spacing. Data on weather, leaf yellowing incidence, crop growth, yield components and grain yield were collected. Rainfall was 280 mm during the cropping season which was poorly distributed, therefore supplementary irrigation was done. Average temperature was 24^{o}C with mean relative humidity of 86.5%.

Disease index results indicated that incidence of rice leaf yellowing disease was high in the unfertilized control plots and low or absent in fertilized plots. Varieties varied significantly ($P<0.01$) between growth parameters and yields components, Saro and Supa had high yield and low yield was obtained in Mbawambili. Fertilizer types also significantly ($P<0.05$) affected rice grain yield. Urea with the rice yield of 1.7 t/ha was the best followed by compost and cow dung manure (1.5 and 1.6t/ha respectively) and the lowest 1.3t/ha was recorded in the control plots. Regression and correlation analyses showed positive correlation between growth components and yield of rice. Generally, Mwangaza and Supa showed outstanding yield performance which should be supplemented by using integrated soil fertility approaches to increase rice productivity.

4.2 INTRODUCTION

Rice (*Oryza sativa* L.) is affected by many biotic diseases caused by fungi, bacteria, viruses, nematodes, and Mycoplasma (Lamo *et al.,* 2015). The incidence of diseases and the damage they cause to rice crops are reported to be increasing wherever the crop is grown (Zouzou *et al.,* 2008). Host plant resistance and an integrated approach to crop management including appropriate control of plant pathogens could reduce much of the disease problem. However, in developing countries, such a strategy is rarely applied effectively. One main reason for the non- effective application of the control measures is the apparent lack of adequate knowledge of etiology and incidence of the diseases.

Despite the high demand for rice worldwide and Africa in particular, the productivity of the crop has remained low in most rice producing countries in Africa countries including Tanzania. In Tanzania rice yield on smallholder farmers' fields range from 1.0 - 2.2 tons ha^{-1} compared to the yield of 5.3 - 6.0 t ha^{-1} and 5.0 t ha^{-1} reported in the rice-growing countries of Vietnam and China, respectively (FAO, 2015). Thus, there is a potential for increasing rice productivity in Tanzania. The climatic conditions like temperature, rainfall, relative humidity, solar radiation; soil types and nutrient status may affect the crop development and performance (De Datta, 1981). A number of biotic and abiotic stresses have been reported to contribute to the low yield of rice in Africa of which poor soil fertility, low yielding cultivars, insect pests, and diseases are the most important (Lamo *et al.*, 2015).

Organic products are highly in demand especially in today's world with concern for consumer's health and ecosystem sustainability (Dubey and Dubey, 2009). Soil fertility is diminish gradually due to various factors such as soil erosion, loss of soil nutrients, water logging and continues cultivation of fields without fallowing. In organic farming various agronomic practices are advised to improve fertility such as fallowing, intercropping, use of organic wastes and bio-fertilizers, as alternate sources to meet nutrient requirements of crops. WARDA (2008) indicated that lowland rice varieties major problems are nitrogen deficiency, extreme temperature, poor water control, disease i.e. bacterial leaf blight, salinity and acidity/alkalinity.

Fertilizer is one of the vital inputs which enhance the increase in yield of rice. Nutrients uptake differs from one rice variety to another. In rice and most field crops, nitrogen is known as the key nutrient, it is one of the most important and essential nutrient. It influences the growth, development, yield and quality of rice (Haefele *et al.*, 2006). Another limiting nutrient which affects rice productivity is phosphorus, which plays important role in cell division, seed formation, root growth, crop maturation and development. Potassium affects grain filling. Zinc as a micronutrient also may limit rice productivity when deficiency. It affects seedlings and it may cause serious problems in rice (Janaki, 2006).

Shemahonge (2016) reported that most small holders' farmers do rarely use fertilizer either inorganic or organic due to various constraints such as financial, availability of fertilizers, bulkiness and transportation. Commonly organic fertilizers used are compost, crop residues or farmyard manure (Kajiru, 2006). Applications of organic materials such as livestock manure and crop residues have been found to bring about a gradual improvement in soil productivity enhance root growth and nutrient uptake and crop performance (Siavoshi, 2011).

4.3 MATERIALS AND METHODS

4.3.1 Description of the study site

The study was carried out at Mingo village (Lubungo ward) in Mvomero District from May to September 2019. The village is located at Latitude: 6.83° South, longitude 37.5° East and elevation of 500 m above the sea level. Based on 30 years weather data (Meteoblue, 2020), the village experience a bimodal rainfall pattern with the short rains falling between late-October to the end of January and the long rains from February to May; a dry season is experienced between June to mid-October. The wards receive monthly precipitation ranging from 6 to 44 mm with a mean monthly minimum and maximum air temperature range of 17°C to 21°C and 26°C to 30°C, respectively. The village was selected for this study because of its long history of rice production and the high incidence of yellowing and stunting problem reported by farmers.

4.3.2 Experimental materials

Planting materials used in the study were four rice varieties; Mbawambili, Mwangaza, Supa, and Saro, which are grown at various areas in Morogoro region. The varieties were collected from farmer's paddy harvest of 2018 cropping season except for Mwangaza and Saro which were collected from the Department of Crop Science and Horticulture at Sokoine University of Agriculture (SUA). Among the selected varieties, Supa and Mwangaza are short duration varieties, while the rest are long duration varieties. Two types of fertilizers used were, Inorganic (Urea 46% N) and Organic (Compost and Cow dung manure). Cow dung manure was collected from farmers' animal sheds (boma) in Lubungo ward and compost was collected from the Horticulture Unit at SUA.

4.3.3 Land preparation, agronomic practices and experimental design

4.3.3.1 Land preparation

Land preparation was done as described by Kanyeka *et al.* (2007) that included ploughing and harrowing using an oxen drawn implement. Levelling was done by using hand hoe to create a favourable condition for planting and emergence of seedlings. Blocking was done across the slope to check erosion and to minimize any incidence of soil fertility gradient that may exist on the field.

4.3.3.2 Other agronomic practices

Other agronomic/management practices after planting was done as recommended for rice. Thinning was done to two plants per hill at 16 days after planting. Weeding was done two times at 21 and 45 Days after planting (DPA). Supplementary irrigation was applied throughout the season so as to rescue crop from drought. Small threat from birds was controlled manually by scarring– off the birds and cattle. Diseases occurrence was scored by counting diseased plant in each of the plots as one of the activities.

4.3.3.3 Experimental Design

The study was laid out as a split-split plot experiment in a Randomized Complete Block Design (RCBD) with four replications and 16 treatment combinations (Kuelh, 2000). Main treatments were four rice varieties that included (i) Mbawambili (ii) Mwangaza (iii) Supa and (iv) Saro. Direct seeding was done by the dibbling method whereby 5 –8 seeds per hill were sown (Kanyeka *et al*., 2007). The sub treatments were different fertilizer types at their recommended rates (i) Cow dung manure at the rate of 5t ha^{-1}, (ii) urea 46% N at a rate of 80 kg N ha^{-1} and (iii) compost at the rate of 5t ha^{-1} (iv) No fertilizer (control). Cow dung manure and compost were applied by broadcasting and incorporated in the soil before sowing, while urea was applied as top dressing in two splits; first at vegetative stage [i.e. 21 Days after planting (DAP)] and second application was done at panicle initiation i.e. 65 DAP

(Kanyeka *et al*., 2007). The experiment covered an area of 196 m^2, whereby the main plot size was 8 m^2 (4 m x 2 m) and subplot area 2 m^2 (2 m x 1 m).

4.3.4 Data Collection

4.3.4.1 Weather data

Rainfall (mm), minimum and maximum temperature (ºC) and percentage relative humidity (RH %) were recorded on daily basis from the Tanzania Meteorology Agency (TMA) situated at SUA.

4.3.4.2 Crop vegetative growth phase

Crop vegetative growth and development stages from emergence to panicle initiation including number of days to 50% emergence, number of days to first tiller, and total number of tillers were recorded (Wopereis *et al*., 2009). Number of days to 50% emergence was determined by observing and counting plants when 50% of them emerged (with two leaves) from sowing to 15 DAP. Number of days to first tiller was recorded when tillering was first noticed on 50% of plants. Number of tillers on five randomly picked plants in each replicate was counted and recorded in all emerging shoots in the hill from the time of planting to when 50% of the plants reached flowering stage. Plant height on five randomly picked plants in each replicate was measured from the ground level to the highest tip of the stem using a ruler and the average was computed.

4.3.4.3 Reproduction phase

Number of days to 50% flowering was recorded when 50% of the plants in a plot had flowered using five plant samples from each plot (Wopereis *et al*., 2009).

4.3.4.4 Ripening phase

Data for ripening phase were collected from flowering stage to maturity stage (Gomez, 1972), where number of panicles on five randomly picked plants was recorded from the centre of each plot and the average number of panicle was computed. Total number of spikelets was recorded from the middle panicle and the average was computed. A total of 1000 seeds from five randomly picked mature rice plants were weighed using an electronic balance to obtain the average weight. The total number of filled and unfilled grains obtained by threshing middle panicles was used to compute the percentage of unfilled grains as presented in Equation 1. Rice harvested from one meter square area in the middle of each plot and threshed accordingly and used to estimate grain yield. The paddy were then adjusted at 14% moisture content using the formula described in Equation 2, and then the grain weight for each plot was recorded and converted in to kg ha^{-1} (Gomez, 1972).

Adjusted grain weight= (A×W) ………………………………………………….. (1)

Adjustment coefficient computed by A= $\frac{100-M}{86}$ …………………………….

……. .(2)

Where A is adjustment coefficient, M is the moisture content (%) of the harvested grains and W is the weight of the harvested grains.

Total dry matter accumulation was determined using the method described by (Gomez, 1972). At physiological maturity, plants from 1 m^2 area in each plot were harvested at ground level, oven dried at 70ºC for 72 hours and then weighed to get dry matter accumulation per plot. Harvest index computed as described by (Fageria, 2001) using Equation 3.

$$Harvest\,index = \frac{Grain\,yield\left(g/m2\right)}{Total\,biomass\left(g/m2\right)} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(3)}$$

4.3.4.4 Leaf yellowing disease assessment

The disease scoring was done 21 DAP, the severity of the syndrome was recorded by observing all the plants in each treatment separately, using a 0-5 scale as per IRRI (2002).

Disease Index was calculated as:

$$DI = \frac{n(3) + n(5) + n(7) + n(9)}{tn}$$

Where: n (3), n (5), n (7), and n (9) = number of plants showing a reaction in a scale (3),(5), (7), and (9) respectively.

tn = total number of plants scored

The resulting DI was classified as:

DI reaction class

0-3 Resistant/tolerant

4-6 Moderate

7-9 Susceptible

4.3.5 Data analysis

Data collected was subjected to the analysis of variance (ANOVA) using Genstat 16^{th} edition software (VSN International). Mean separation test was conducted using Turkey's Honest Significance Difference Test at $P \leq 0.05$ as the level of significance. A regression analysis was calculated using Pearson product moment correlation coefficient (r) in the Microsoft office-Excel programme to explore the relationship between growth and yield components of rice grown at Mingo village in Mvomero district of Tanzania.

4.4 RESULTS

4.4.1 Weather information of the study area during experiment

Weather information of the study area during growing season was as indicated in Figure 4.1, the recorded mean temperature was 24^0 C, highest maximum mean temperature was 31^0C and the lowest was 27^0C. Relative humidity ranged from 91% which was the highest to 80% the lowest. Mean rainfall amount recorded high in April (273.2 mm) and low in between June-September (8 mm).

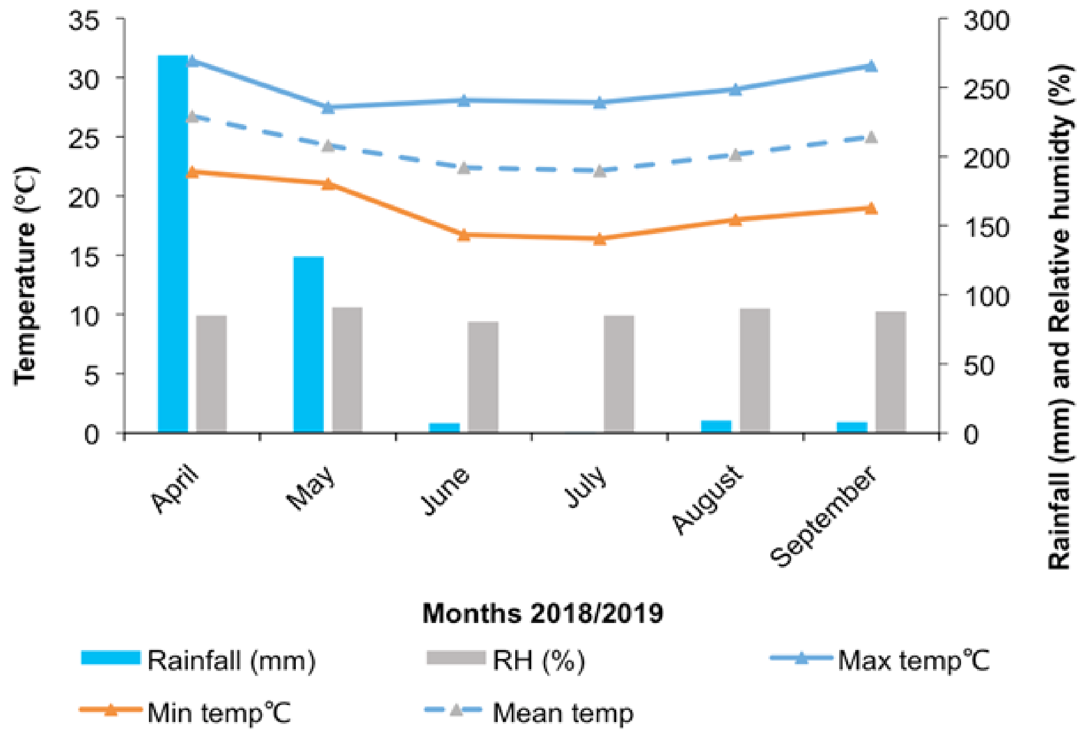

Figure 4.1: Mean values for rainfall, Relative humidity, maximum and minimum Temperature

4.4.2 Severity of leaf yellowing of rice grown in Mingo village

Disease index (DI) was calculated to combine disease incidence and severity as per IRRI (2002). Disease index for leaf yellowing was significant between treatments (Table 4.1). Highest DI was recorded in Mbawambili while the lowest was in Mwangaza .There was no significant difference between types of fertilizers, significantly ($p<0.001$), variation occurred between fertilized plots and unfertilized plots, and highest disease index was observed in unfertilized plots. Between all combinations Mbawambili and control had the highest disease index (1.95) followed by Saro and control (1.35), then Supa and control (0.6) and the least was in Mwangaza and control (0.45).

Table 4.1: Severity of leaf yellowing of rice grown in Mingo village

Main factor (Rice variety)	Yellowing (DI)	Interaction (Rice variety x Fertilizer type)	Yellowing (DI)
Mbawambili	0.48b	Mbawambili x Compost	0.0a
Mwangaza	0.11a	Mbawambili x Control	1.95c
Saro	0.33ab	Mbawambili x Cow dung	0.0a
Supa	0.15a	Mbawambili x Urea	0.0a
Mean	0.272	Mwangaza x Compost	0.0a
SE±	0.082	Mwangaza x Control	0.45a
CV%	120.1	Mwangaza x Cow dung	0.0a
p-value	0.007	Mwangaza x Urea	0.0a
		Saro x Compost	0.0a
		Saro x Control	1.35bc
		Saro x Cow dung	0.0a
Sub factor (Fertilizer type)		Saro x Urea	0.0a
Compost	0.0a	Supa x Compost	0.0a
Control	1.088b	Supa x Control	0.6ab
Cow dung	0.0a	Supa x Cow dung	0.0a
Urea	0.0a	Supa x Urea	0.0a
Mean	0.272	Mean	0.272
SE±	0.0816	SE±	0.1633
CV%	120.1	CV%	120.1
p-value	<0.001	*p*-value	<0.001

***DI = Disease Index (Incidence and Severity of yellowing symptom)

4.4.3 Effects of fertilizer type and rice varieties on growth, yield and yield components of rice

Results indicated that some of the growth parameter such as days to 50% emergence and flowering, plant height, number of tiller per plant, number of panicles, panicle weight were significantly different ($p<0.01$) (Table 4.2). Number of spikelet's were not significant different between rice varieties. Highest number of days to emergence was observed in Saro variety (13 days) and the lowest was Mwangaza variety (8 days). In days to 50% flowering, the highest was observed for Supa while Mwangaza had the lowest, and the same occurred for panicle weight where Supa had the highest (4.3 g) and Mwangaza had the lowest (3.3g).

It was observed that days to 50% emergence, flowering, plant height, number of tillers, panicle, spikelet's, and panicle weight were significantly affected by the type of fertilizer used (Table 4.2) .

Table 4.2: Effects of fertilizer type and rice varieties on growth components of rice grown at Mingo village

	Days to 50% emergence	**Days to 50% flowering**	**Plant height (cm)**	**Number of tillers/ plant**	**Number of panicles**	**Panicles weight (g)**	**Number of spikelets**
Main factor (Rice variety)							
Mbawambili	11.3c	74.9b	53.9a	14.4ab	158.7ab	4.2c	23.0a
Mwangaza	8.0a	73.2a	60.9b	11.6a	158.2a	3.3a	22.6a
Saro	13.75d	74.9b	53.2a	15.6b	157.6a	3.8b	23.3a
Supa	10.0b	75.1b	55.2ab	13.8ab	160.1b	4.3d	23.3a
Mean	10.8	74.5	55.8	13.8	158.6	3.9	23.1
SE±	0.281	0.1852	1.798	0.816	0.495	0.0294	0.212
CV%	10.5	1.0	12.9	23.6	1.2	3.0	3.7
p-value	<0.01	<0.01	0.017	0.011	0.007	<.001	0.19
Sub factor (Fertilizer type)							
Compost	11.2b	70.9b	52.6b	12.2ab	157.1a	3.6a	21.6b
Control	12.3c	79.4d	39.4a	9.8a	157.2a	3.7ab	19.1a
Cow dung	10.3b	70.1a	59.9c	14.8b	157.1a	3.8b	22.8c
Urea	9.2a	77.7c	71.3d	18.7c	163.1b	4.4c	28.9d
Mean	10.8	74.5	55.8	13.8	158.6	3.9	23.1
SE±	0.281	0.1852	1.798	0.816	0.495	0.0294	0.212
CV%	10.5	1.0	12.9	23.6	1.2	3.0	3.7
p-value	<0.01	<0.01	<0.001	<0.001	<0.001	<0.001	<0.001

Means bearing the same letter(s) within the column are not significantly ($p > 0.05$) different according to Turkey's Honest Significance Test. CV = Coefficient of Variation and SE± = Standard error of means.

There were significant differences in percentage of filled and unfilled grains and total dry matter between treatments. Grain weight (1000 seeds), grain yield and harvest index did not differ significantly between rice varieties (Table 4.3). On the other hand, fertilizer types significantly (p<0.001) affected grain weight (1000 seeds), percent of filled and filled grains and total dry matter.

Table 4.3: Effects of fertilizer type and rice varieties on yield and yield components of rice grown at Mingo village

	Grain weight (1000 seeds)	Filled grains (%)	Unfilled grains (%)	Grain Yield (t/ ha)	Total dry matter (g/ m^2)	Harvest index
Main factor (Rice variety)						
Mbawambili	25.4a	81.1ab	18.9ab	1.5a	458.2ab	0.3a
Mwangaza	25.2a	80.7a	19.3b	1.5a	458.6ab	0.3a
Saro	25.0a	81.1ab	18.9ab	1.5a	459.3b	0.3a
Supa	25.2a	81.5b	18.5a	1.5a	457.8a	0.3a
Mean	25.2	81.1	18.9	1.5348	458.47	0.34
SE±	0.1769	0.18	0.18	0.0092	0.355	0.002
CV%	2.8	0.9	3.8	2.4	0.3	2.4
p-value	0.45	0.057	0.057	0.826	0.03	0.805
Sub factor (Fertilizer type)						
Compost	23.5b	81.3b	18.7b	1.5b	456b	0.3b
Control	22.8a	77.6a	22.4c	1.3a	347.7a	0.3d
Cow dung	24.8c	81.7b	18.34b	1.6c	462.9c	0.3c
Urea	29.7d	83.9c	16.1a	1.7d	567.2d	0.3a
Mean	25.2	81.1	18.9	1.5	458.5	0.3
SE±	0.1769	0.1811	0.1811	0.0092	0.355	0.002
CV%	2.8	0.9	3.8	2.4	0.3	2.4
p-value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Means bearing the same letter(s) within the column are not significantly (p > 0.05) different according to Turkey's Honest Significance Test. CV = coefficient of variation and SE± = Standard error of means.

4.4.4 Interaction effects of fertilizer type and rice varieties on growth, yield and yield components of rice grown.

Interaction effects of fertilizer type and rice varieties on growth components of rice varied significantly (p<0.01) in all treatments (Table 4.4). Highly significant difference between treatments was observed in number of days to 50% flowering and number of spikelet's as shown in Figure (4.2 and 4.3 respectively). Combination of Saro and control treatment showed highest number of days to 50% emergence (15.3) while Mwangaza and Urea showed the lowest number of days (6.5). In days to 50 % flowering Mwangaza and Control showed highest number of days (79.8) and the lowest was observed in Supa and Cow dung combination (69.8). Plant height varied significantly among treatments, the mean value was 55.8 cm, Supa and Urea recorded highest height plants (70.7) while Mwangaza and control had the lowest (37.9).

Number of tillers per plant was also affected by fertilizer type used against rice variety. Results indicated that combination of Mbawambili and Urea had the highest (19.0) while the lowest (8.0) was recorded in Mwangaza and control. A combination of Supa x Urea and Mwangaza x Urea recorded highest number of panicles (163.8) and the lowest was recorded in Mwangaza and cow dung (155.0). For panicle weight Mwangaza x compost and Mwangaza x control did not differ significantly but they recorded the lowest weight (3.0g) while a combination of Saro x Urea and Saro x cow dung recorded the highest mean weight (4.2g). Number of spikelet's was affected, a combination of Supa and Urea showed highest number (29.3) and Mbawambili and control showed the lowest (19.0), the mean value was 23.1 (Table 4.4).

Table 4.4: Interaction effects of fertilizer type and rice varieties on growth components of rice grown at Mingo village

Interaction (Rice variety x Fertilizer type)	**Days to 50% emergence**	**Days to 50% flowering**	**Plant height (cm)**	**Number of tillers/plant**	**Number of panicles**	**Panicles weight (g)**	**Number of spikelets**
Mbawambili x Compost	11.3cde	71.8b	49.9abc	13.5abcd	157.0a	3.9cdef	21.8de
Mbawambili x Control	13.0efg	79.8ef	39.1a	10.0ab	157.0a	4.0defg	19.0ab
Mbawambili x Cow dung	11.3cde	70.0ab	58.0bcde	15.3abcd	158.0ab	4.1efg	23.0de
Mbawambili x Urea	9.5bcd	78.3de	68.5def	19.0cd	162.8bc	4.6h	28.3f
Mwangaza x Compost	8.5abc	70.0ab	60.3cdef	9.8ab	155.2a	3.0a	21.0bcd
Mwangaza x Control	9.5bcd	77.8d	37.9a	8.0a	158.8abc	3.0a	19.3ab
Mwangaza x Cow dung	7.5ab	70.0ab	66.7cdef	12.3abc	155.0a	3.1a	21.5cd
Mwangaza x Urea	6.5a	75.0c	78.6f	16.5bcd	163.8c	3.8cde	29.3f
Saro x Compost	14.5fg	71.0ab	50.8abcd	13.3abcd	156.2a	3.5b	22.0de
Saro x Control	15.3g	79.5def	39.5a	10.8abc	156.0a	3.7bcd	19.5abc
Saro x Cow dung	13.0efg	70.8ab	55.2abcde	17.0bcd	155.8a	3.7bc	22.8de
Saro x Urea	12.3def	78.5de	67.4cdef	21.3d	162.2bc	4.2fg	29.0f
Supa x Compost	10.5cde	70.8ab	49.5abc	12.3abc	159.8abc	4.2fg	21.8de
Supa x Control	11.5de	80.8f	41.0ab	10.3ab	157.0a	4.1efg	18.5a
Supa x Cow dung	9.5bcd	69.8a	59.8cde	14.5abcd	159.8abc	4.3g	23.8e
Supa x Urea	8.5abc	79.0ab	70.7ef	18.0bcd	163.8c	4.7h	29.3f
Mean	10.75	74.5	55.8	13.8	158.6	3.9	23.1
SE±	0.563	0.3703	3.597	1.632	0.991	0.0588	0.424
CV%	10.5	1.0	12.9	23.6	1.2	3.0	3.7
p-value	0.986	<0.01	0.658	0.997	0.068	0.079	0.042

Means bearing the same letter (s) within the column are not significantly ($p > 0.05$) different according to Turkey's Honest Significance Test. CV = coefficient of variation and SE± = Standard error of means.

Yield and yield components were also affected by treatments significantly for all parameters as shown in Table 4.5. Grain weight of 1000 seeds for treatments showed that combination of Mbawambili x Urea had the highest grain weight (30.5gm) and Supa x control had the lowest (22.7gm). Percent of filled and unfilled grains was highest (84.1 and 33.9) in Mbawambili x Urea and Mwangaza x control, and lowest (77.2 and 15.9) in Mwangaza x control and Mbawambili x Urea respectively.

Grain yield varied significantly between treatments. Mean value were 1.53t/ha. The highest mean yield were obtained in a combination of Supa x Urea (1.7t/ha) and the lowest were obtained in a combination of Mbawambili x control and Supa x control. Highest total dry matter were obtained in a combination of Mwangaza x Urea (567.8 g/m^2) while the lowest were obtained in a combination of Mwangaza x control ($347g/m^2$).

Harvest index varies significantly among treatments. Mean value was 0.34, the highest value were recorded in a combination of Mbawambili x control, Mwangaza x control, Saro x control, Supa x control respectively, other treatments had the H.I of 0.3.

Table 4.5: Interaction effects of fertilizer type and rice varieties on yield ad yield components of rice grown at Mingo village

Interaction (Rice variety x Fertilizer type)	Grain weight (1000 seeds)	Filled grains (%)	Unfilled grains (%)	Grain Yield (t/ ha)	Total dry matter (g/ m^2)	Harvest index
Mbawambili x Compost	23.4abc	80.9b	19.1c	1.5bcd	455.8b	0.3cd
Mbawambili x Control	22.9ab	78.0a	21.9d	1.3a	347.8b	0.4e
Mbawambili x Cowdung	24.9c	81.6b	18.4c	1.6bcd	462.5a	0.3cd
Mbawambili x Urea	30.5d	84.1d	15.9a	1.8e	566.8e	0.3ab
Mwangaza x Compost	23.8abc	80.8b	19.2c	1.5b	456.5b	0.3abc
Mwangaza x Control	22.8a	77.2a	22.9d	1.4a	347a	0.4e
Mwangaza x Cowdung	24.7bc	81.2b	18.9c	1.6d	463cd	0.3d
Mwangaza x Urea	29.4d	83.8cd	16.2ab	1.8e	567.8e	0.3ab
Saro x Compost	23.3abc	81.3b	18.8c	1.5bcd	456.8b	0.3abcd
Saro x Control	22.8a	77.6a	22.5d	1.4a	348.5a	0.4e
Saro x Cowdung	24.6bc	81.5b	18.5c	1.6cd	465.0d	0.3cd
Saro x Urea	29.5d	83.9d	16.1a	1.8e	567.0e	0.3ab
Supa x Compost	23.8abc	82.1bc	17.9bc	1.5bc	455.0b	0.3bcd
Supa x Control	22.7a	77.6a	22.4d	1.3a	347.5a	0.4e
Supa x Cowdung	24.9c	82.4bcd	17.6abc	1.6cd	461.2c	0.3d
Supa x Urea	29.4d	83.8cd	16.2ab	1.7e	567.5e	0.3a
Mean	25.2	81.1	18.9	1.5348	458.47	0.34
SE±	0.3538	0.3623	0.3623	0.01841	0.71	0.004
CV%	2.8	0.9	3.8	2.4	0.3	2.4
p-value	0.618	0.438	0.438	0.677	0.247	0.458

Means bearing the same letter(s) within the column are not significantly ($p > 0.05$) different according to Turkey's Honest Significance Test. CV = coefficient of variation and SE± = Standard error of means.

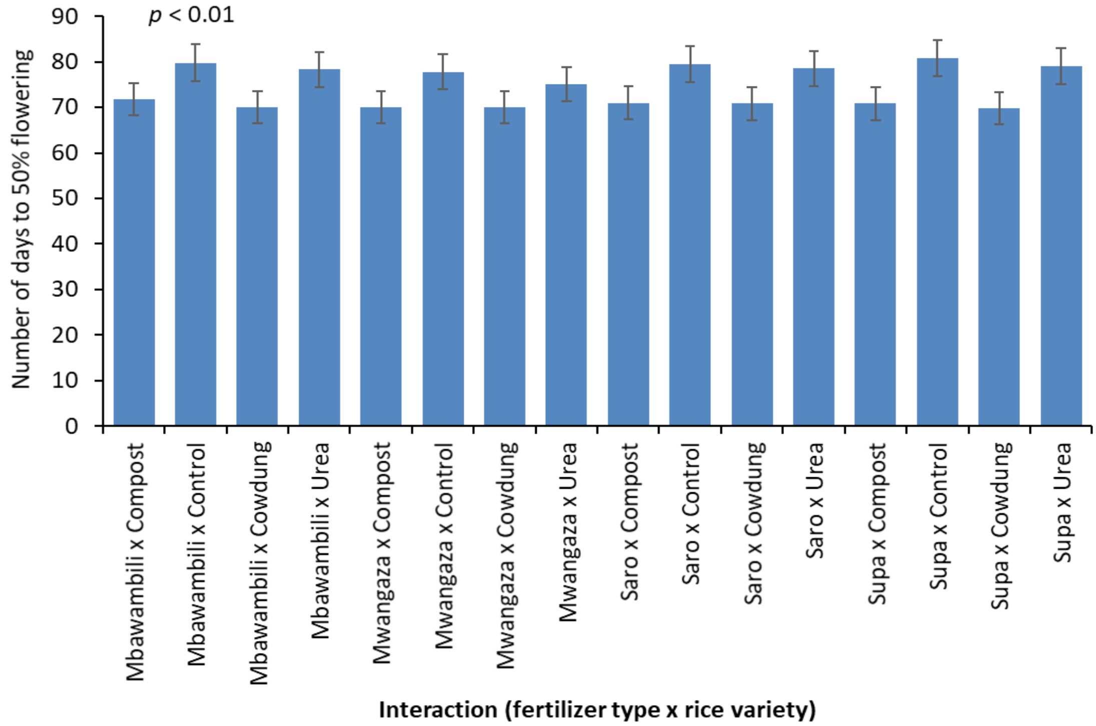

Figure 4.2: Interaction effect of fertilizer type and rice varieties on number of days to 50% flowering

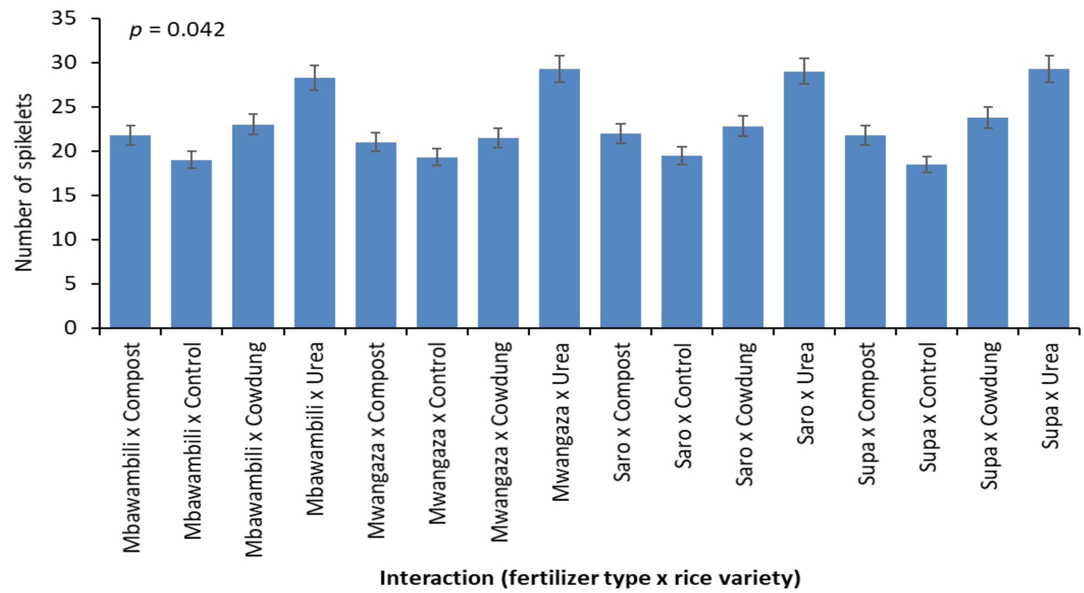

Figure 4.3: Interaction effects of fertilizer type and rice varieties on number of spikelet's

4.4.5 Relationship between growth components and yield of rice grown at Mingo village due to the effects of applied treatments

Based on regression and correlation analysis, results (Table 4.6) indicate that most of growth components had significant relationship with yield of rice grown at Mingo village due to the effects of applied treatments. The plant height (cm), number of tillers per plant, number of panicles, panicles weight (g), number of spikelets, grain weight of 1000 seeds (g), filled grains (%), total dry matter (g/m^2) was significantly ($p<0.05$) higher, indicating positive correlation with rice yield. The number of days to 50% emergence (r = -0.44, R^2 = 0.19, $p < 0.0003$), percentage unfilled grains (r = -0.9, R^2 = 0.8, $p < 0.05$), and harvest index (r = -0.8, R^2 = 0.6, $p < 0.05$) showed a significant negative correlation with rice yield. However, the relationship between number of days to 50% flowering and rice yield due to the effects of applied treatments was not significant (r = -0.1, R^2 = 0.01, $p < 0.4$).

Table 4.6: Correlation (r) between growth and yield components of rice grown at Mingo village

	Days to 50% emergence	**Days to 50% flowering**	**plant height (cm)**	**number of tillers/plant**	**Number of panicles**	**Panicles weight (g)**	**Number of spikelets**	**Grain weight of 1000 seeds(g)**	**Filled grains (%)**	**Unfilled grains (%)**	**Total dry matter (g/ m^2)**	**Harvest index**
Grain Yield (t/ ha)												
R	0.4**	0.1*	0.8**	0.7**	0.6**	0.4**	0.9**	0.9**	0.9**	0.9**	1.0**	0.8**
R^2	0.2	0.01	0.7	0.5	0.4	0.2	0.9	0.8	0.8	0.8	0.9	0.6
I	22.4	78.8	-62.6	-19.4	136.9	1.6	-12.4	0.2	59.5	40.5	-299.2	0.6
M	-7.6	-2.8	77.2	21.7	14.2	1.5	23.1	16.3	14.1	-14.1	493.7	-0.2
SE	2.0	3.6	6.7	2.8	2.3	0.4	1.2	1.1	0.9	0.9	19.2	0.02
P	3.1 E-3	0.44	5.0E-17	1.8E-10	8.7E-8	4.1E-3	8.8E-28	2.9E-22	9.2E-23	9.2E-23	1.02E-34	6.1E-14

** = Significant ($p<0.05$) relationship with rice yield. * = Relationship not significant ($p>0.05$). r = Correlation, R^2 = Regression, I = Intercept, M = Slope.

4.5 DISCUSSION

4.5.1 Weather during the growing season

Rice is grown under so diverse soil and climatic conditions, primarily it is a tropical and subtropical crop. From this study the recorded mean temperature was 24^0 C, highest maximum mean temperature was 31^0C and the lowest was 27^0C. The average temperature required throughout the life period of the crop ranges from 21 to 37º C. The temperature sum, range, distribution pattern, and diurnal changes, or a combination of these may be highly correlated with grain yields. The climatic conditions like temperature, rainfall, relative humidity, solar radiation; soil types and nutrient status may affect the crop development and performance (Shemahonge, 2016). Rice crop needs a hot and humid climate. It is best suited to regions which have high humidity, prolonged sunshine and an assured supply of water. Each growth phases and every stage has an optimum temperature range for its proper development. Duration of the critical temperature, have a great impact on physiological status of the plant. Ran *et al.* (2018) reported that, temperature-related indicators (maximum temperature and minimum temperature) had more significant impact on rice yield than rainfall.

Mean rainfall was recorded high in April (273.2 mm) and low between June-September (8 mm). Rice crop requires more water than any other crop. As a result, rice cultivation is done only in those areas where minimum rainfall is 115 mm, average annual rainfall between 175—300 mm are the most suitable. Supplementary irrigation was done throughout the growing season because it was off season.

Relative humidity from data collected ranged from 91% which was the highest to 80% the lowest. It directly influences the water relations of rice plant and indirectly affects leaf growth, photosynthesis, pollination, occurrence of fungal diseases such as rice blast and leaf blast and finally economic yield. It reduces dry matter production in rice plant and leaf water potential. Reduced transpiration influences translocation of food materials and nutrients, moderately high RH of 60 to 70% is beneficial to rice crop development and performance (Oikeh *et al.*, 2008).

4.5.2 Severity of leaf yellowing of rice grown in Mingo village

The extents of variation among the treatments in respect of disease index are as presented. Disease index (DI) was calculated to combine disease incidence and severity as per IRRI (2002). Disease index for leaf yellowing was significant among treatments at ($p<0.001$), Mbawambili recorded highest DI within varieties, while Mwangaza had the lowest. This disease index was calculated based on the assumption of yellowing leaf disease but in the experiment no any incidence of disease was recorded. This yellowing might therefore be associated with nutrient deficiency as it occurs in control plots or quality of variety used. Mbawambili recorded high incidence of yellowing because it is a local variety and it is highly affected by nutrient deficiency. Mwangaza which is an early maturing variety showed resistance to various diseases including yellowing syndrome.

4.5.3 Effects of fertilizer type and rice varieties on growth, yield and yield components of rice

Growth parameters of rice were significantly influenced by varieties and type of fertilizer used. Mwangaza variety which is the early maturing had few numbers of days to 50% emergence and flowering, with few numbers of tillers per plant and lowest number of panicle weight. Differential genetic characters and production potential which might have led to differential response to nutrient management practice leading to variation in growth parameters .Early emergence and synchronized tillering, being dependent on the mineral nutrition and hereditary characters of the crop determine the tiller numbers and tiller emergence in later growth stages (Choudhary *et al.,* 2007). Myint *et al. (*2010) associate increase of SPAD value in all treatments with varietal characteristic rather than soil fertility.

Chemical fertilizer offers nutrients which are readily soluble in soil solution and thereby rapidly available to plants. Application of Urea influenced almost all parameters and varies significantly among treatments. Days to 50% emergence, plant height, number of tillers, number of panicles, panicle weight and number of spikelets were high in plots applied with Urea, except for number of days to 50% flowering. Siavoshi (2011), reported similar results in his study where plant height, number of tillers per hill, spikelet number per panicle ,grain yield and 1000-grain weight were influence by the application of organic and chemical fertilizers. Huang *et al*. (2011) reported that application of nitrogen at early stages of rice growth increase number of panicles per unit area.

Compost and cow dung had almost similar performance in all parameters with exception of number of tillers per plant. Muhammad *et al*. (2008) observed similar results with application of organic manure and compost in rice. Plots with no fertilizer recorded lowest means in all parameters and there was a delay in crop emergence and flowering, this might be due to the fact that nutrient availability from organic sources is influenced by microbial action and improved physical condition of soil as reported by Sarker *et al*. (2004).

Grain yield components are highly influenced by Environmental conditions (Katsura *et al.*, 2008). There was no varietal difference in grain weight grain yield and harvest index. 1000-grain weight is a stable varietal characteristics, size of the grain cannot grow to a size greater than that permitted by the hull (Asmamaw, 2017). Varieties significantly influenced percent of filled and unfilled grains and total dry matter. All varieties showed averaged grain yield (1.5 t/ha) although Mbawambili, Saro and Supa had similar percent of filled grains. The type of fertilizer used influenced yield and yield components of rice at Mingo village. Plots applied with Urea indicated high grain weight (1000 seeds), percent of filled and lowest unfilled grains percentage, highest mean grain yield (1.7 t/ha) and total dry matter (567.2). Similar results were obtained by Siavoshi (2011) and Salem (2006).

Performance of rice under compost and cow dung manure varied significantly in all parameters although performance in each parameter was almost the same. Lowest mean grain yield was obtained in control plots with highest percentage of unfilled grains and lowest percent of filled grains. This is due to fact that filled grains are the ones influencing grain yield of crop. Total dry matter was also influenced by type of fertilizer used; highest total dry matter was observed in Urea and lowest in control plots. Total dry matter indicates the amount of biomass accumulated during vegetative as influenced by nutrients and climatic condition. Urea fertilizer influenced high biomass accumulation hence high total dry matter, where plots with no fertilizer had low biomass production. A report by Myint *et al*. (2010) show that rice yield and N availability of low fertility soil increased within three years of continuous organic manure application. However, it may depend on the kind and amount of organic matter used.

4.5.4 Interaction effects of fertilizer types and rice varieties on yield and yield components of rice grown at Mingo village

Interaction between two factors varied significantly in growth components of rice at Mingo village. There was a variation among parameters recorded. Combination of Mwangaza and Urea influenced number of days to 50% emergence which has the lowest number of days. Supa and Cow dung combinations showed least number of days to 50% flowering while Supa and Control showed longest number of days to 505 flowering. Plant height was influenced by treatments combinations where shortest plant was observed in Mwangaza and control plots and tallest plant were observed in combination of Mwangaza and Urea. In his study (Kolleh, 2017) reported Mwangaza to be the tallest under various weed management practices.

Tillering is an important trait for grain production and is thereby an important aspect in rice yield. Hasanuzzaman *et al*. (2010) reported increase in number of tillers in rice plants due to influence of different fertilizer combinations. Number of tillers per plant was observed to be few in Mwangaza x control combination while Saro and Urea had the highest number of tillers. Supa x Urea combination had the highest number of panicle per area and fewest in Mwangaza and Cow dung. The same was observed for number of spikelets in combination of Supa and Urea and the lowest in Supa and Combination of Mwangaza x compost and Mwangaza x control had the lowest panicle weight while Supa x Urea showed highest number of panicle weight Studies reported that higher yield is generally achieved either by increasing panicles per unit area (Huang *et al.*, 2011). Furthermore, studies reports that organic sources offer more balanced nutrition to the plants, especially micro nutrients which positively affect number of tiller in plants (Siavoshi, 2011).

Not all the grains present in the panicle were filled. Number of unfilled grains in a panicle depends on genotype, growing environment and the nutrient provided (Sangita *et al.,* 2018). Significantly higher filled and low filled grain/panicle was due to the application of Urea in Mbawambili variety as compared to control plot.

Mean grain weight treatment combinations was 25.2 and combination of Supa x Urea Mwangaza x Urea with showed highest number of grain weight, while Supa and control combination gives out lowest grain weight .The increase in grain yield could be due to the increase in yield components. In tillering crops like rice, the number of tillers per unit area, the plant height and leaf size are the physical components influencing dry matter production during different time intervals (Harish *et al.*, 2017). Grain yield was low in combination of Mbawambili, Supa and control while Mbawambili, Saro and Urea have the highest grain yield. These results are in line with those of Phongpan *et al. (*1993) who reported that grain yields were significantly increased with Nitrogen treatment over the control. The improvement in rice yield potential might come from increased the biomass production as per Zhao *et al.(*2020).

4.6 CONCLUSION

Generally, weather condition at Mvomero district is suitable for rice production as this study and other reports indicated. Occurrence of yellowing and rice stunting was low in this study as compared to reports of farmers except, few incidence of yellowing were observed and scored in control plots in all treatments. Among varieties used Mwangaza showed good performance in almost all growth and yield parameters while Mbawambili had the poor performance. Urea, among all fertilizers, showed outstanding performance in yield whilist compost and cow dung had similar average performance which did not differ significantly. Impact of types of organic fertilizer used was not significant especially in yield components due to soil fertility status of Mingo, and the time taken to complete the experiment. These results are therefore considered preliminary as the experiment was done only once due to time restriction.

It is hereby recommended that farmers at Mingo should start using integrated ways for fertility increase in their fields. Organic fertilizer should be applied in a right/ recommended amount and supplement with fallowing, planting leguminous crops after rice in order to increase fertility. Furthermore, studies to assess effectiveness of concentrated foliar fertilizer using organic materials should be conducted. This may give out potential output and help reduce bulkiness of organic fertilizer, easy solubility of nutrients, portability, cost and environmental friendly practice.

Mbawambili which is a common and local variety used does not show good results as compared to Supa, Mwangaza and Saro and therefore adaptation of these varieties in the area is vital to increase productivity.

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CHAPTER FIVE

5.0 CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

Generally, lack of information, and knowledge on the cause of yellowing syndrome were the main reasons limiting the effective management of rice yellowing problem. The use of susceptible rice varieties and poor agronomic practices were additional constraints to management of yellowing syndrome. Our study has provided a base-line data defining the current status of farmers' knowledge on rice yellowing and stunting of plants. This information can be used to initiate a process to identify the abiotic and/or biotic causal agent(s) of rice yellowing and stunting of plants, which will in turn be the basis for developing appropriate management measures and improve rice yield.

The current study has revealed the presence of several seed borne pathogenic fungi associated with rice seed samples from Mingo and Kimambila villages in Mvomero district and indicates the possibility of disease episode when such infected seeds are used to raise the next rice crop. In as much as the results of this study may be considered preliminary, they nevertheless imply fungi associated with rice seeds are potential threat to its production.

Availability of nitrogen in both sites is very low and it is the major cause of leaf yellowing syndrome which in turn results to plants stunting and low yield of rice. Continuous rice monoculture cropping system has result to decline in soil fertility due to deterioration in physical properties of the soil like texture and microbial existence. To improve the situation, growing rice after a dual purpose grain legume in the field can improve nitrogen content and influence organic carbon content hence improve rice plant health

Phosphorus and Potassium where sufficient but this might be for short term production and after a time they will be depleted hence the need to practice fallowing and leaving rice straws in the field after harvesting.

Generally, weather condition at Mvomero district is suitable for rice production as this study and other reports indicated. Yellowing syndrome from suspected diseases were not observed in the experimental area, although yellowing occurred in plots with control treatments and it was due to low amount of nitrogen. Among varieties used Mwangaza showed good performance in almost all growth and yield parameters while Mbawambili had the poor performance. Urea, among all fertilizers showed outstanding performance in yield, this is due to solubility of nutrients to the crop and availability of N in the fertilizer.

Compost and cow dung had similar average performance which did not differ significantly. Impact of types of organic fertilizer used was not significant especially in yield components due to soil fertility status of Mingo, and time taken to complete the experiment .The quality of fertilizer used might be affected by the kind of feeds consumed by the animal or material used in making compost.

Organic fertilizers that can be used are in form of compost, crop residue and farmyard manure but farmers don’t have enough knowledge on how to use them. Transportation problem due to bulkiness and high cost due to large requirement per unit area and its availability are the limitations to uses of these fertilizers.

5.2 Recommendations

This study recommends that farmers at Mingo should start using integrated nutrient management strategies to increase fertility in their fields. Organic fertilizer should be applied in a right recommended amount and supplement with planting leguminous crops after rice in order to increase fertility, also supplement with industrial N in a balanced proportion. Soybeans, chick pea and green grams have shown a lot of potential in alleviating the problem of soil fertility. Such legumes can be cultivated during off-season at the time the land used to lie fallow.

A study to assess efficiency and effectiveness of concentrated foliar organic fertilizers is considered necessary in order to solve challenges of bulkiness, cost, availability and environmental toxicity which are associated with the current types of fertilizers used.
Organic fertilizers may also be used following recommended rates and types as peer soil requirements. UREA, DAP, Calcium of Ammonium nitrate, Sulphate of Ammonia, Minjingu mazao and Minjingu nafaka are among recommended inorganic fertilizers that are used in Tanzania.

Mwangaza and Saro varieties are among high yielding and early matured varieties and they should therefore be adapted in order to increase productivity.
Based on this results a further study into the importance of these seed borne pathogens with respect to rice growth and yield is warranted for effective and sustainable management of rice seed heath and seed quality. Farmers should however be trained on improved cultivation methods through demonstration, farmer field schools and also be encouraged to practice the technology that is being recommended.

APPENDICES

Appendix 1: Questionnaire for data collection from rice growers in Mvomero district

Code no. ………….. Date …………………….

A. DETAILS ABOUT GROWER, LOCATION AND EDUCATION.

a) Sex …………….…… Age…………..…..
b) Size of the farm …………………..
c) Village…………….Ward…………….Province ………………..
d) Education level…………..……..

B. RICE PRODUCTION/ CROPPING SYSTEM

1. What kind of production/ cropping system is utilized on your farm?
   (a) Mixed cropping (b) Sole cropping ☐
2. If it is mixed cropping, outline the crops grown together with rice? ………………………………………………………………..………………………

C. IMPORTANCE OF RICE IN THE LOCAL PRODUCTION SYSTEM

3. How long since you started growing rice?
   (a) 1 year (b) 3 – 5 years (c) Over 5 years ☐
4. Compared to other crops you are also producing, to what extent does rice contribute to your income?
   (a) High (b) Low (c) Medium ☐
5. How much rice (in kg) do you harvest per season? …………..…………….…….…...…...
6. Normally, 1kg of rice is sold at what price ……………………………..……….
7. The rice you harvest, what do you use for? ☐
   (a) Commercial (b) Home consumption

D. CULTURAL PRACTICES AND VARIETIES GROWN

8. What management practices do you apply to your rice crop?

(a) Fertilization (b) Pruning (c) Irrigation (d) Weeding

(e) Others …………………………………...

9. What variety of rice do you grow?

……………………..………………………………………………………..……

E. RICE PRODUCTION CONSTRAINTS

10. What constraints do you face in rice production?

……………………………………………………………………………….………

11. If mentioned in (10) above, for how long have you been experiencing yellowing and stunting of rice? ....

12. Which variety is most affected by yellowing and stunting of rice?

…………………………………..…………………………………………..……

13. During which season and stage does leaf yellowing and stunting of rice occur...

14. What measures have you taken so far on management of leaf yellowing and stunting of rice.

……………………………………………………………………….……………...

15. How effective have been the measures taken?

……………………………………………………………………….…….………...

16. Any other information about the problem…………………………………….……….