

**EVALUATION OF ADVANCED KENYAN WHEAT GENOTYPES FOR RESISTANCE
TO STEM RUST (*Puccinia graminis* f.sp. *tritici*) RACES**



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**A Thesis Submitted to the Graduate School in Partial Fulfillment for the Requirements of
Master of Science in Agronomy (Crop Protection) of Egerton University**

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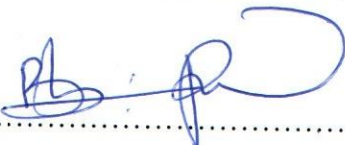
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DEDICATION

This thesis is dedicated to my late mother Mrs. Ettah Kabarika Tenge and sister Rose Nekesa Tenge.

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ABSTRACT

Stem rust caused by a fungus race *Ug99* (*Puccinia graminis* f.sp.*tritici*) is an important disease of wheat in Kenya. The races of *Ug99* occur widely in Kenya and due to its virulence nature wheat cultivars released usually lose their resistance within a short period of time. The aim of the study was to evaluate advanced Kenyan wheat genotypes for resistance to stem rust both at seedling and adult plant stages. For resistance an experiment was carried out in the greenhouse. A total of fifty genotypes were used, forty five advanced genotypes and five local checks for both adult and seedling stage resistance. Scoring of disease at the seedling stage was done following the Stakmans scale while the modified Cobb scale was used for field evaluation. The seedling stage experiment identified genotypes KSL 50, 31, 33, 54, 51, 156, 81 and 44 as being very resistant. The field experiment when using the Final Disease Severity (FDS) identified genotypes KSL 142, 71, 144, 31 and 44 as having high resistance levels. The area under disease progress curve was calculated for each genotype. The disease assessment using incidence and severity established Mau-Narok as having 32.1% and 41.4%. Kabatini had 7.9% and 23.3% Njoro reported no disease. Mau-Narok reported high disease occurrence through the AUDPC values. Farming practices revealed that spraying regimes, varieties used, seed source and rotation influenced disease levels. The growers with certified seed, two or three sprays and rotations had low or no disease. Use of resistant varieties and development of resistance in genotypes is paramount to management and control of stem rust. The use of resistant genotypes combined with one or two sprays is sufficient for management of stem rust.

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LIST OF ABBREVIATIONS AND ACRONYMS

ANOVA	Analysis of Variance
AUDPC	Area Under Disease Progress Curve
ASALs	Arid and Semi-Arid Lands
BGRI	Borlaug Global Rust Initiative
CI	Coefficient of Infection
CIMMYT	International Maize and Wheat Improvement Center
CIMMYT	Centro Internacional de Mejoramiento de Maíz
CV_i	Coefficient of variation of each genotype
EAAPP	Eastern Africa agricultural productivity project
EPZA	Export processing zone authority
EU	European Union
FAO	Food and Agriculture Organization
FDS	Final Disease Severity
GoK	Government of Kenya
ICARDA	International Centre for Agricultural Research in the Dry Areas
IFPRI	International Food Policy Research Institute.
KALRO	Kenya Agriculture and Livestock Organization
KARI	Kenya agricultural Research Institute
KIPPRA	Kenya Institute for Public Policy Research and Analysis
LSD	Least Significant Difference
M.A.S.L	Meters above Sea Level
MALF	Ministry of Agriculture Livestock and Fisheries
MOA	Ministry of agriculture

SAS/STAT	Statistical Analysis Software
SHF	Small Holder Farmers
S_i	Standard deviation of a genotype across environments
S²	The variance of a genotype across environments
TTKSK	Race of <i>Ug99</i>
TTKST	A new variant of race <i>Ug99</i>
<i>Ug99</i>	TTKSK
USAID	United States Agency for International Development
USDA	United States Department of Agriculture

CHAPTER ONE

1.0 INTRODUCTION

1.1 BACKGROUND INFORMATION

Wheat is a key cereal crop for global food security (Mondal, Rutkoski, Velu, Crespo-Herrera, Guzmán, Bhavani, Lan, He & Singh, 2016). Multiple abiotic challenges due to climate change and evolving pathogen and pests pose a major concern for increasing wheat production globally (Mondal, Rutkoski, Velu, Crespo-Herrera, Guzmán, Bhavani, Lan, He & Singh, 2016). They are an ever-present threat to wheat and are among the most virulent (Dubin & Brennan, 2009). The rusts of wheat can be disseminated thousands of kilometers across continents and oceans by wind (Kolmer, 2005). Cereal rusts are heteroecious and macrocyclic requiring two taxonomically unrelated hosts to complete a five spore stage life cycle (Kolmer, 2013). Stem or black rust has historically caused severe losses to wheat production worldwide (Njau, Keller, Macharia, Singh & Wanyera, 2009). Wheat is susceptible to three types of rust; stem (black) rust (*Puccinia graminis* f.sp. *tritici* race *Ug99*), leaf (brown) rust *Puccinia triticina*, and stripe (yellow) rust *Puccinia striiformis* f.sp. *tritici* (Dubin & Brennan, 2009).

Wheat stem rust is one of the most destructive disease of durum and common or bread wheat (Olivera, Badebo, Xu, Klindworth & Yue, 2012). The causal race, commonly known as *Ug99* and designated as TTKS based on the North American nomenclature, carries virulence for several genes commonly present in wheat germplasm. All Kenyan germplasm are known to be susceptible or partially susceptible to *Ug99* although no proper documentation has been done (Njau, Keller, Macharia, Singh & Wanyera, 2009). In most wheat-growing regions of the world, existing environmental conditions favour stem rust infection, which could lead to epidemic buildup (Singh, Hodson, Huerta- Espino, Jin, Bhavani, Njau, Herrera-Foessel, Singh & Govindan, 2011). Knowing pests and diseases that may cause injuries and are likely to affect plant health and quality is critical to minimizing the gap between attainable yield and actual yield (Duveiller, Singh & Nicolas, 2007). With world population increasing and food security

projected to become more critical, increasing wheat yield potential in the developing world remains a high priority (Duveiller, Singh & Nicolas, 2007).

Agriculture production in Kenya has not kept rapidity with population growth rate and the country has become a net importer of its two major staple foods, maize and wheat (Mohajan, 2013). Wheat is the second most important staple food in Kenya, which accounted for 17% of staple food consumption. Recently in urban areas of Kenya use of wheat and rice is increasing (Mohajan, 2013). Though wheat is not nearly as widely grown as maize or rice, it is an important component of the country's domestic food production – being grown on about 4 percent of the country's arable land 160,000 hectares out of 4,000,000 hectares of arable land (Soko directory, 2016). Kenya faces challenges in wheat production which are the impact of climate change, Land degradation and persistent biotic and abiotic stresses (Macauley, 2015). Rapid population growth associated with difficulty in meeting the projected demand for food. Poor mechanization, inadequate or weak policy environment. Dwindling financial resources needed for Research and Development (Macauley, 2015). The demand for wheat products has consistently increased over the last five years leading to an increase in wheat imports (MALF, 2015). Domestic wheat consumption increased from 671,000 tonnes in 2004 to 1,850,000 tonnes in 2014. Despite the fact that the consumption rate is high, its production seems to be lower than it is expected (Soko directory, 2016).

1.2 Statement of the problem

Stem rust disease of wheat has increased in the recent past due to new races resulting from mutation especially the *Ug99* group. The new races have overcome the resistant genes making wheat as a crop very vulnerable to stem rust. Long term growing of susceptible varieties may result to increased amount of inocula in the fields. This is as a result of favorable weather patterns mainly found in the tropics. Stem rust disease causes grain yield losses making it impossible to attain maximum yields per unit area of land. In Kenya such losses have resulted in deficit of production which has caused the country to import large amounts of wheat due to the higher demand that exceeds the supply. Production of wheat is not sustainable because farmers spray several times to obtain a clean crop. Environmental concerns on chemical use and affordability by resource poor farmers is a major constraint.

Disease incidence and severity varies from place to place and one season to another due to the differential weather patterns. Disease incidence and severity are affected by agronomic practices during farming which differs from one region to another. Wheat stem rust disease occurrence in percent incidence and severity is related to control and management practices. Efficient control and management practices are limited and this is compounded by breakdown of resistant varieties after a short time and mutation of the rust pathogen, leading to a high turnover of varieties released to the farmers.

1.3 Objectives

1.3.1 General Objectives

To enhance wheat production and food security in Kenya through integrating better rust disease management approach

1.3.2 Specific Objectives

1. To determine seedling and adult stage resistance of the advanced Kenyan wheat lines to stem rust.
2. To determine the differences in stem rust disease intensity using incidence and severity across the sites.

1.4 Null Hypotheses

1. Advanced Kenyan wheat lines do not possess seedling and adult stage resistance to stem rust.
2. The intensity of stem rust disease does not vary across the environments

1.5 Justification

Genetic improvement for resistance is one of the most sustainable methods for managing stem rust disease, due to its cost effectiveness and being environmentally friendly in management of stem rust. Aggressive new races of stem rust being discovered are due to the large quantities of inocula always present. The races are the main causes of stem rust epidemics in the major growing regions. Planting of resistant varieties reduces the disease pressure. Although resistant genotypes have been released over time, virulence of the rust pathogen has resulted in susceptibility leading to high turnover of the new varieties. Therefore new varieties

which possess resistance to emergent races of stem rust, superior in performance with high resistance levels and grain yield are required for durable breeding purposes. The varieties should be released to farmers for improved productivity.

Disease incidence and severity are parameters that are important in determining percentage of stem rust disease occurrence and levels. They are necessary in providing an understanding of the causes of stem rust disease occurrences and epidemics in the major wheat growing regions. The two parameters are important in decision making for establishing management practices needed for effective control. Assessment of stem rust disease is thus important for the development of a reliable and effective management strategy. It is a guide to the use of integrated management of the stem rust disease. Control measures need to be constantly developed for success in stem rust disease control.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Wheat Production in Kenya

Kenya's wheat production is less than one quarter of its annual demand, and the deficit is offset by imports. The bulk of the wheat imports are from Russia, Ukraine, Lithuania, Estonia, Germany, Poland, and Australia (GAIN, 2016). Wheat growing areas in Kenya include the scenic Rift Valley regions of Uasin Gishu, Narok, Marakwet, Elgeyo, Londiani, Molo, Nakuru and Timau areas. These areas have altitudes ranging between 1200 m and 1,500 m above sea level, with annual rainfall varying between 800 mm and 2,000 mm, with up to 2,500 mm on higher grounds (EPZA, 2005). The main growing regions have been the areas above 1500 m in the Nakuru, Uasin Gishu, Trans Nzoia and Laikipia counties. The break-up of some of the large farms in these Counties resulted in a switch to maize production or a combination in which wheat is grown as a cash crop and maize is produced for subsistence consumption (FAO, 2013). The crop is grown largely for commercial purposes on a large scale. Kenya is self-sufficient in the hard variety of wheat, but is a net importer of the softer variety (EPZA, 2005).

Small scale farmers grow wheat in small areas of less than 5 acres while large scale farmers grow the crop on more than 5 acres of land. Furthermore, large-scale farmers are more mechanized in wheat production compared to small-scale farmers. The large-scale farmers dominate wheat production with a share of 75 % of the wheat area and 83 % of production (Nyangito, Ikiara & Ronge, 2002). Domestic wheat accounts for less than 40 % of the total consumption with the rest being met from imports. Like in the case of maize, millers prefer the imported wheat because it is at times cheaper and is of superior quality than the local wheat. To protect the producers, wheat imports are taxed (Nyoro, Wanzala & Awour, 2001).

2.1.1 Wheat Production Practices in Kenya

The production system in Kenya includes varieties grown, use of certified and non-certified fungicide use and cropping systems. The commonly used fungicides in Kenya are same as ones found in North America belonging to two major classes these are the Strobilurins and

triazoles (Wegulo, Stevens, Zwingman & Baenziger, 2012). Strobilurins, with a chemistry based on a natural product from a mushroom, are fungicide of new generation and proved to be quite effective, protective, eradicator and potential broad-spectrum substances against foliar diseases of winter wheat (Gaurilčikienė, 2010). The triazoles chemical family of fungicides was introduced in the 1980s, which consists of numerous members: difenoconazole, myclobutanil, propiconazole, tebuconazole, tetraconazole, triadimefon, and triticonazole. They are important tools against diseases of turf grasses, vegetables, citrus, field crops and ornamental plants (Rouabhi, 2010).

Crop rotation is one of the most important means of managing disease in small grains. It is found also in major wheat growing regions of Kenya of a wheat/maize or wheat/legume which are the common beans, peas and also tomatoes and potatoes are used. Farmers worldwide have rotated different crops on their land for many centuries. This agronomic practice was developed to produce higher grain yields by replenishing soil nutrients and breaking disease and pest cycles (EU, 2012). The yields of wheat experienced in Kenya range from 24.8-30.7 bags per hectare (MALF, 2015). The commonly grown wheat varieties found in Kenya are NjoroBWII, Robin, Mwamba, Duma, Kwale, Korongo, Eagle10, Heroe, Ngamia and Farasi. The levels of resistance to stem rust disease in the varieties have reduced.

2.2 Life Cycle of Stem rust Wheat Disease

Rusts are important pathogens of angiosperms and gymnosperms including cereal crops and forest trees. With respect to cereals, rust fungi are among the most important pathogens (Kolmer, 2013). Cereal rusts are heteroecious and macrocyclic requiring two taxonomically unrelated hosts to complete a five spore stage life cycle (Kolmer, 2013). The fungus is an obligate parasite, heteroecious and has five spore stages (Leonard & Szabo, 2005).

Stem rust fungi is an obligate parasite in nature, requiring a living host tissue for growth and reproduction (Schumann & Leonard, 2000). The disease cycle of the rust pathogen starts when the susceptible wheat crop gets exposed to the stem rust spores, urediniospores. As the host plant matures, the urediniospores produce teliospores (Schumann & Leonard, 2000). With rains and favorable temperatures, the teliospores germinate and produce basidiospores borne on structures called basidia (Leonard and Szabo, 2005). Basidiospores then infect the alternate hosts such as common barberry, germinate, and produce a haploid mycelium, which colonizes the leaf tissue, which form pycnia inside the leaf (Leonard and Szabo, 2005). The pycnia produce

receptive hyphae and pycniospores of a single mating type that serve as the female and male gametes (Schumann & Leonard, 2000). Mating of the male and female pycniospores results in the formation of aeciospores that are dicaryotic (N+N) and produced in aecia on the lower surface of the leaf 7 to 10 days post fertilization (Roelfs *et al.*, 1992). Aeciospores are then hydrospectically released from aecia and are airborne to infect wheat meters or even kilometers away resulting in production of dicaryotic (N+N) uredinia with urediniospores under optimum temperatures of 30°C and a dew period of six to eight hours. This completes the life cycle (Schumann & Leonard, 2000).

In the absence of barberry or other alternate hosts, urediniospores are the only functional spores in the disease cycle of stem rust (Schumann & Leonard, 2000). In tropical and subtropical climates, mycelium and urediniospores on volunteer wheat and noncrop grass hosts begin epidemics (Schumann & Leonard, 2000).

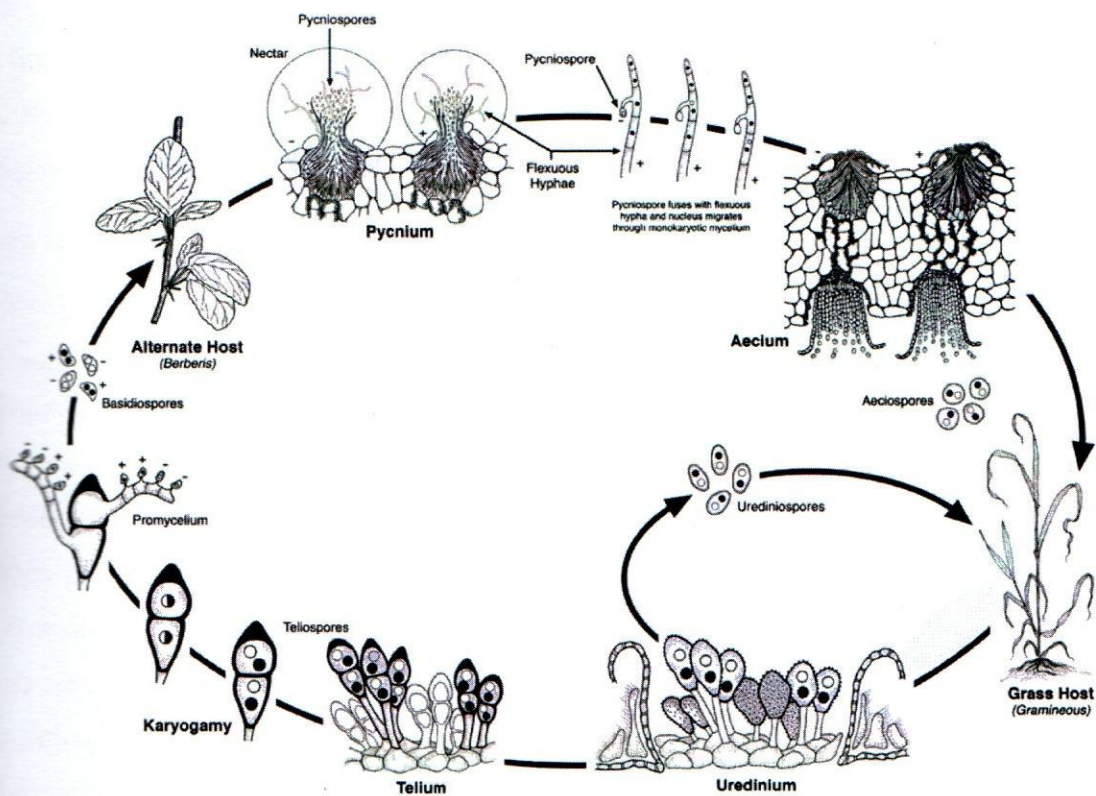


Plate 2.1: Life cycle of wheat stem rust

2.2.1 Wheat Stem rust Race Ug99

Race *Ug99*, or TTKSK was first identified in Uganda in 1998 and has been recognized as a major threat to wheat production. Its spread in 2006 to Yemen and Sudan and further spread towards North Africa, Middle East and West-South Asia is predicted -aided by predominant wind currents and large areas of wheat varieties that are susceptible and grown under environments favourable for survival and multiplication of the pathogen (Singh, Hodson, Huerta-Espino, Jin, Njau, Wanyera, Herrera-Foessel & Ward, 2008). Stem rust is one of the most serious diseases of bread and durum wheat worldwide. The discovery of new stem rust races in Africa, *Ug99* and its variants, brings a new threat to global wheat production. Currently, the research of stem rust in wheat is focusing on identifying further resistance genes to control *Ug99* and its derivatives (Haile & Roder, 2013). Stem rust (race *Ug99*) belongs to one of several *formae speciales* in *P. graminis*. It appears as elongated blister-like pustules, or uredinia, most frequently on the leaf sheaths of a wheat plant, but also on true stem tissues, leaves, glumes, and awns. Stem rust pustules on leaves develop mostly on the lower side, but may penetrate and produce limited sporulation on the upper side (Singh, Hodson, Huerta-Espino, Jin, Njau, Wanyera, Herrera-Foessel & Ward, 2008). Stem rust is generally considered warm temperature rust (Eversmeyer & Kramer, 2000).

The *Ug99* group of races of the stem rust fungus is widely recognized as a threat to wheat production worldwide because of the races' fast evolution and migration, and to the susceptibility of wheat varieties grown on over 90 % of the world's wheat area (Singh, Hodson, Huerta-Espino, Jin, Bhavani, Njau, Herrera-Foessel, Singh & Govindan, 2011). Race TTKSK possesses broad virulence, especially virulence to genes commonly used in combinations for stem rust resistance in wheat cultivars (Jin & Singh, 2006). The stem rust resistance gene *Sr31* derived from rye has been used as an important source of stem rust resistance in many wheat cultivars worldwide. Isolates of stem rust with virulence to *Sr31* were identified from Uganda in 1999, while stem rust susceptibility in wheat lines with *Sr31* was observed in Kenya in 2003 and 2004. An isolate collected from Uganda in 1999 and an isolate collected from Kenya in 2004, identified to be race TTKSK (Jin & Singh, 2006).

The stem rust resistance gene *Sr36* confers a near-immune resistance reaction to many races of Stem rust and is highly effective against race TTKSK which possesses unusually broad virulence combinations. Because this gene is widely used in United States on soft winter wheat

germplasm and cultivars, it has been considered to be an important source of resistance to TTKSK (Jin, Szabo, Rouse, Fetch, Pretorius, Wanyera & Njau, 2009). The emergence of virulence on *Sr24* within the TTKSK race cluster has increased the vulnerability of wheat to stem rust worldwide because of the widespread use of this gene in breeding (Jin, Szabo, Pretorius, Singh, Ward & Fetch 2008). Rain favours disease by depositing them on the plants, and increasing the humidity (Roelfs, Singh & Saari, 1992).

2.2.2 Factors that Determine Stem rust Development

The amount of disease that develops in a plant community is determined by the host, the pathogen and the environment and can be depicted in the form of a disease triangle. A fourth factor, namely 'human interference' making a disease square can be added, but, as the other three aspects have a degree of human influence, the disease triangle is sufficient as a framework for discussing the various factors that affect disease (Keane & Kerr, 1997). The amount of disease that develops is often determined by the pathogenicity of the prevalent population of the pathogen. The term pathogenicity comprises both the virulence of the pathogen that is its ability to infect cultivars which have certain resistance genes and its aggressiveness which is the vigour that it infects cultivars without resistance genes or cultivars whose resistance genes are not activated by the pathogen (Keane & Kerr, 1997). Disease epidemics result from the combination of inoculum, favourable environment, and host susceptibility (Duveiller, Singh & Nicolas, 2007).

The main host factor affecting disease development is the occurrence of cultivars in the host population that are susceptible to the particular pathogen. For a disease epidemic to occur, the host plant population must be largely susceptible to attack by the pathotypes of the pathogen in the vicinity (Keane & Kerr, 1997). Environmental factors have traditionally been considered to have the major impact on disease development. Even if a susceptible host and a virulent pathogen are present in a certain locality, a common situation when the farmer has no choice but to plant the particular host, serious disease will not occur unless the environment favours its development (Keane & Kerr, 1997). Disease development depends on environment as well as genes in the host and pathogen (Shaner & Finney, 1977). Incidence data are frequently collected in epidemiological studies of plant disease because they provide a convenient and useful assessment of disease intensity. However, the characteristics of disease incidence data should be taken into account for analysis (Madden & Hughes, 1995).

2.3 Management of Wheat stem Rust

Wheat rusts can be controlled worldwide by planting resistant varieties of wheat which is more sustainable. Although fungicides may be effective against wheat rust, they are not economically feasible. Fungicides are only recommended when based on accurate monitoring data and as an emergency control measure until resistant wheat varieties are again available (FAO, 2008).

2.3.1 Breeding for Resistance to Wheat stem Rust Disease

The long-term success of breeding for disease resistance is influenced by the following factors; the nature of the pathogen and diversity of virulence in the population, diversity and type of genetic resistance, screening methodology and selection environment for tracking resistance (FAO, 2002). Genetic studies have suggested that wheat genotypes that are resistant to a given rust disease in many locations, as indicated by low average coefficients of infection, often contain multiple major or minor genes for resistance (FAO, 2002). Genetic resistance, rather than fungicide use, has been and will continue to be the primary means of combating rust disease of wheat in developing countries. In developed countries, the demand by consumers for food produced without pesticide application will increase the need for disease resistant wheat varieties (Shiferaw, Smale, Braun, Duveiller, Reynolds & Muricho, 2013).

The breeding of disease resistant wheat varieties is the chief line of long-term defense for wheat crops against stripe rust, and in fact, for all rust diseases (ICARDA, 2011). The collaborative international effort that successfully developed rust resistance in wheat had a tremendous impact on world food supplies. It is estimated that modern rust-resistant wheat varieties account for about 30 % of the increase in wheat production worldwide, with consequent benefits for food production, poverty reduction, and food security. These varieties now account for 95 % of the wheat in developing countries (Dubin & Brennan, 2009). The best long-term strategy to mitigate the threat from *Ug99* is to identify resistant sources among existing materials, or develop resistant wheat varieties that can adapt to the prevalent environments in countries under high risk, and release them after proper testing while simultaneously multiplying the seed (Singh, Hodson, Huerta-Espino, Jin, Njau, Wanyera, Herrera-Foessel & Ward 2008).

The alien resistance gene *Sr31* has been used in agriculture on the largest scale since 1980s in spring, facultative and winter wheat breeding programmes worldwide except Australia. Its use in CIMMYT wheat improvement resulted in the release of several popular cultivars worldwide (Singh, Hodson, Huerta-Espino, Jin, Njau, Wanyera, Herrera-Foessel & Ward 2008).

Many wheat cultivars throughout the world have the stem rust resistance gene *Sr31*, which was introgressed into wheat on a translocated chromosomal fragment from rye. This gene has provided highly effective resistance for many years (Kolmer, 2005). Some old Kenyan varieties were found to have adult plant resistance probably due to the presence of non-race specific gene *Sr2* complex which among others can be exploited in breeding for resistance in Kenyan wheat (Njau, Keller, Macharia, Singh & Wanyera, 2009). The stem rust resistance gene *Sr24* is effective against most races of stem rust, including race TTKSK and is used widely in commercial wheat cultivars worldwide (Jin, Szabo, Pretorius, Singh, Ward & Fetch 2008).

Implicit in CIMMYT's mandate to help produce additional food in a sustainable manner in the Third World is the development of wheat germplasm to achieve this end (Dubin & Rajaram, 1996). The CIMMYT Bread Wheat Breeding Programme is attempting to thwart epidemics due to well known pathogens globally through gene accumulation and gene deployment (FAO, 2002). The future of global food security in wheat depends on new varieties and management practices to meet the demand from differentiated value chains, address the projected negative impacts of climate change, and reverse the stagnating productivity trends in the post-Green Revolution era (Shiferaw, Smale, Braun, Duveiller, Reynolds & Muricho, 2013). The early maturing cultivars were introduced to permit a second crop or to avoid flowering and grain filling during hot weather. The early maturing cultivars escape much of the damage caused by stem rust by avoiding the growth period of the fungus. The widespread use of resistant cultivars worldwide has reduced the disease as a significant factor in production (Roelfs, Singh & Sauri, 1992).

2.3.2 Cropping Patterns and Alternate Varieties

Diversified cropping of wheat by avoiding the sowing of mega-varieties across large cropped areas is another possible defense strategy against wheat rust. In most areas of the Middle East, East Africa and South Asia, farmers have been planting the same varieties for 20-30 years (ICARDA, 2011). Cultivars have been developed over the last few decades that often mature

prior to development of severe stem rust epidemics. The early maturity of the crop shortens the number of generations available for the development of either leaf or stem rust epidemics, and allows the crop to escape serious damage. Unless overwintering of inocula has occurred, the generation time for rust is significantly shortened and the disease is not able to reach the economic loss threshold (Eversmeyer & Kramer, 2000). A gene deployment strategy is used to increase diversity in the wheat population in a given region. Use of gene deployment over a wide area will increase the probability of inocula landing on an incompatible host and, thus, negate a large amount of the potential inocula. Early gene deployment was a result of less interchange of germplasm among the wheat programmes (Eversmeyer & Kramer, 2000).

2.4 Impact of Stem rust to Wheat Production

The frequently encountered vulnerability of monogenic resistance to stem rust requires dedicated breeding for durable resistance in wheat (Jin, Szabo, Rouse, Fetch, Pretorius, Wanyera & Njau, 2009). Aggressive new strains of wheat rust diseases – stem rust and stripe rust – have decimated wheat yields in recent harvests. Key areas affected are East Africa, North Africa, the Middle East, Central Asia and the Caucasus. Rust diseases have reduced the wheat harvests in Egypt, Ethiopia, Iran, Kenya, Morocco, Syria, Turkey, Uzbekistan, and Yemen, in the past five years (ICARDA, 2011). Because of the susceptibility of 90 % of the wheat varieties grown worldwide, the *Ug99* group of races was recognized as a major threat to wheat production and food security. The spread, either wind-mediated or human-aided, to other countries in Africa, Asia, and beyond is evident (Singh, Hodson, Huerta- Espino, Jin, Bhavani, Njau, Herrera-Estrella, Singh & Govindan, 2011). Because rust continually evolves to overcome existing genetic resistance, no form of resistance lasts forever. Today, a new threat from wheat rust looms (Dubin & Brennan, 2009). The fungicides used for the control of wheat leaf diseases have increased costs of production, because of the multiple applications required to protect the crop before it matures (Wanyera, Kilonzo & Macharia, 2010).

Many farmers in these tropical highland areas of Africa work under subsistence conditions. Wheat diseases in tropical regions can be severe and require significant efforts to control. For economic and environmental reasons, host plant resistance is the most appropriate and sustainable disease control method for economic and environmental reasons (Dubin & Rajaram, 1996). Stem rust epidemics are causing grain losses of up to 70 % in experimental plots

and over 70 % in farmers' fields. This is yield of sprayed versus unsprayed wheat crop. Spraying only reduces but does not eliminate the disease. It is therefore possible to get yield losses higher than this when relative to a clean crop. In the year 2007, farmers who never controlled the disease at all, lost 100 % of their crop regardless of the variety (Wanyera, 2008).

Kenya has had to rely on wheat imports to meet the domestic and regional demand for wheat and wheat products. Increased wheat imports have led to a further decline in wheat production because imports dampen domestic prices, which are a disincentive to production (Nyangito, Ikiara & Ronge, 2002). Stem rust race *Ug99* is responsible for up to 100 % yield loss of wheat (Mwando, Tabu, Otaye & Njau, 2012). Adverse impacts of climate change can also be expected from the likely rise in the spatial distribution and intensity of existing pests, diseases, and weeds, due to higher temperatures and humidity. The magnitude of the overall effect is difficult to assess but it is likely to be highly regionalized (Fact sheet, 2008). Farmers will face the challenge of dealing with increased pest problems, or new pest challenges, within the constraints of what science can provide and within the EU's pesticide authorization regulatory framework (Fact sheet, 2008).

2.5 Constraints of Wheat Production in Kenya

Wheat stem rust is present in Kenya throughout the year and significant quantities of inoculum are always present. This increases the likelihood of developing races with new virulence (Jin, Szabo, Rouse, Fetch, Pretorius, Wanyera & Njau, 2009). The variants found in 2006 and 2007 with virulence to *Sr24* and *Sr36* in the TTKS lineage are globally significant because resistance to TTKSK in many adapted cultivars is conferred by these genes. It is anticipated that additional new variants in the TTKS lineage will develop; thus, monitoring of virulence in the stem rust population in Kenya is needed (Jin, Szabo, Rouse, Fetch, Pretorius, Wanyera & Njau, 2009). Although Kenya has a well-developed agricultural research system, use of modern science and technology in agricultural production is still limited (GoK, 2010). Inadequate research-extension-farmer linkages to facilitate demand-driven research and increased use of improved technologies continue to constrain efforts to increase agricultural productivity (GoK, 2010).

In addition, subdivision of family-owned farms into smaller units for inheritance purposes continues to hinder efficient wheat farming in Kenya (GAIN, 2016). The cost of key

inputs such as seed, pesticides is high for resource-poor farmers. Such high costs lead to low application and adulteration of inputs (GoK, 2010). Apparent consumption has been growing at an average annual rate of over 4 % and shows no sign of slowing. With production largely stagnant, the gap has been met by the elimination of exports in the early 1960s and a continuous increase in imports (FAO, 2013). The demand for consumption rises at an estimated 7% per year, driven by population growth, increased urbanization and changing diets. The annual production is about 350,000 tonnes, yet the demand stands at 750,000 tonnes. This means the local production meets only 40 % of the total consumption, hence Kenya imports 60 % of its wheat requirements (Wanyera, 2008).

Global agriculture is facing the probable impact of global warming. Recent studies suggest that the production of major commodities has declined since 1980 due to global warming (Macauley, 2015). In addition to inherently high climate variability, the looming threat of higher temperatures and more vicious droughts (arising from climate change) is a major concern. Further, high incidences of diseases, insect-pests, and parasitic plants, and sub-optimal soil nitrogen have also presented a continuous challenge to cereal productivity in SSA (Macauley, 2015). The low level of mechanization in African agriculture has continued to serve as a huge impediment towards advancing cereal production, especially of wheat and rice which, in turn, results in the high cost of producing these crops (Macauley, 2015).

2.6 Parameters for Disease Measurement

2.6.1 Incidence, Severity and AUDPC

The area under the disease progress curve (AUDPC) estimates the area under the actual infection curve. It is expressed in %-days (accumulation of daily percent infection values) and interpreted directly without transformation. The higher the AUDPC, the more susceptible is the clone or variety. The AUDPC is calculated from all the three ratings at different time thus leading to a more accurate phenotypic evaluation (Ali, Muneer, Xu, Durrishahwar, Hassan, Ullah, Noor & Ullah, 2012). The AUDPC has a lower error variance than statistics associated with the logit transformation of severity data hence a superior measurement (Shaner & Finney, 1977). An epidemic can be defined as an increase in disease with time or more generally as change in disease with time. It is a dynamic process. The fundamental depiction of an epidemic is the disease progress curve, a plot of disease proportion verses time. The AUDPC and its

various elements express the interaction of pathogen, host and environment over time (Madden, 1980).

The Area under the Disease Progress Curve (AUDPC) can be an efficient instrument to evaluate the epidemic development of foliar pathogen considering each genotype susceptibility and specific architecture (Paraschivu, Cotuna & Paraschivu, 2013). The AUDPC is easily calculated and should be a useful criterion for selection (Shaner & Finney, 1977). Calculation of AUDPC uses a lot of data available and does not obscure the variation in rate of disease development because of transformation (Shaner & Finney, 1977). There is also a correspondence between genotype susceptibility and AUDPC showing that the most susceptible wheat cultivars record higher AUDPC values (Paraschivu, Cotuna & Paraschivu, 2013). Incidence is defined as the proportion (0 to 1) or percentage (0 to 100) of diseased entities within a sampling unit. This rather dry definition provides the needed generality so that, in specific situations, incidence can be the proportion (or percentage) of diseased leaves on a plant (Seem, 1984). Incidence-severity relationships in plant disease have had an important impact on the development of disease assessment methods (Seem, 1984).

2.7 Stability Test for Disease Severity for Resistance of Genotypes to Stem rust

The ability of some crop varieties to perform well over a wide range of environmental conditions has long been appreciated by the agronomist and plant breeder (Finlay & Wilkinson, 1963). One of the main reasons for growing genotypes in a wide range of environment is to estimate their stability (Freeman, 1973). Type one stability occurs when a genotype is stable among-environment and variance is small. Type two occurs when a genotype is stable and response to environments is parallel to the mean response of all genotypes in the trial. Type three occurs when a genotype is stable and the residual coefficient from the regression model on the environmental index is small (Lin, Binns & Lefkovitch, 1986). Edaphic variation between localities and the seasonal variation in any one locality are very great (Finlay & Wilkinson, 1963).

Stability depends on whether they are based on the deviations from the average genotype effect or on the genotype x environment (GE) term, and whether or not they incorporate a regression model on an environmental index (Lin, Binns & Lefkovitch, 1986). The smaller the numerical values of variance for a genotype across environments (S_i^2) and coefficient of

variation of each genotype (CV_i) the more stable is the genotype (Letta & Tilahun, 2007). The variance of a genotype across environments (S_i^2) and Coefficient of variation (CV_i) can be a measure of stability (Lin, Binns & Lefkovitch, 1986). The ideal variety having general adaptability is the one with maximum yield potential in the most favourable environment and maximum phenotypic stability (Finlay & Wilkinson, 1963).

CHAPTER THREE

3.0 ASSESSMENT OF ADVANCED KENYAN SELECTED WHEAT GENOTYPES FOR RESISTANCE TO STEM RUST RACES (*Puccinia graminis* f.sp. *tritici*) IN KENYA

3.1 Abstract

Stem rust (*Puccinia graminis* f.sp. *tritici*) of wheat (*Triticum aestivum*) has caused wheat yield losses in Kenya for years and the trend shows the situation has worsened. The objective of this research was to identify elite genotypes for adult plant and seedling stage resistance. Adult plant resistance study was done under field conditions in three locations. Scoring for disease resistance was done following the modified Cobbs scale. Seedling stage resistance assessment was done in the greenhouse and scored following the Stakmans scale. Genotypes KSL 144, 50, 44, 115 were identified as having the lowest infection levels. Area Under Disease Progress Curve (AUDPC) for each genotype was calculated which revealed KSL 142 (28.1), and 144 (28.1) having low values indicating resistance. The same genotypes performed well with the Final Disease Severity (FDS) values showing resistance. The variance (S_i) and coefficient of variation (CV_i) were calculated from the FDS and yield values, which distinguished stable genotypes. The stable genotypes for disease severity were KSL 69 (8.8%), 161 (14.9%), 54 (12.4%) and 156 (18.24%). The relationship between yield and AUDPC was strong and negative, $r = -0.943$ same as yield and FDS relationship $r = -0.84$. Variation for yield performance was recorded with KSL 137 ($2.63t\ ha^{-1}$) and KSL 31 ($2.52t\ ha^{-1}$) showing high performance. The thousand kernel weight values were not significant for the three location at ($P < 0.05$). The advanced genotypes that consistently performed such as KSL 137, 156, 144 and KSL 142 should be recommended for release as varieties or used in improving local varieties in the Kenyan wheat stem rust breeding programme or potentially in the Eastern Africa region.

3.2 Introduction

Wheat (*Triticum aestivum*) is one of the worlds' most productive and important crops' in the 21st century. Currently there is increased consumption and demand for grain, for fuel as well as food (Curtis & Halford, 2014). Wheat yields must therefore be increased which is an important strategy to prevent food shortages (Curtis & Halford, 2014). It is one of the key staple

crops for global food security, providing more than 35 % of the cereal calorie intake in the developing world, 74 % in the developed world and 41 % globally from direct consumption (Shiferaw, Smale, Braun, Duveiller, Reynolds & Muricho, 2013). Wheat is the second most important cereal staple food after maize in Kenya (USAID, 2010). In Kenya it is mostly grown in the Rift Valley, some areas of upper Central province (Nyandarua, Nyeri) and parts of Meru (Tirau) (USAID, 2010).

In most wheat-growing regions of the world, existing environmental conditions favour stem rust infection, which could lead to epidemic build-up (Singh, Hodson, Huerta- Espino, Jin, Bhowani, Njau, Herrera-Foessel, Singh & Govindan, 2011). An estimated 80-90% of all global wheat cultivars growing in farmer's fields are now susceptible to *Ug99* or variants (*Ug99* factsheet, 2010). *Ug99* is the only known race of wheat stem rust that has virulence for an extremely important resistance gene - *Sr31*. In addition, *Ug99* has virulence against most of the resistance genes of wheat origin and other resistance genes from related species (*Ug99* factsheet, 2010). The stem rust resistance gene *Sr31* derived from rye has been used as an important source of stem rust resistance in many wheat cultivars worldwide. However, isolates of stem rust with virulence to *Sr31* were identified from Uganda in 1999. Similarly stem rust susceptibility in wheat lines with *Sr31* was observed in Kenya in 2003 and 2004 (Jin & Singh, 2006).

Ug99 possess broad virulence, especially virulence to genes commonly used in combinations for stem rust resistance in wheat cultivars (Jin & Singh, 2006; Njau, Keller, Macharia, Singh & Wanyera, 2009). Detection in Kenya of a new variant *TTKST* in 2006 with virulence to gene *Sr24*, which caused severe epidemics in 2007 in some regions of Kenya and rendered about half of the previously known *Ug99*-resistant global wheat materials susceptible, has further increased the vulnerability globally (Singh, Hodson, Huerta-Espino, Jin, Njau, Wanyera, Herrera-Foessel & Ward, 2008). The emergence of virulence on *Sr24* within the *TTKST* race cluster has probably increased the vulnerability of wheat to stem rust worldwide because of the widespread use of this gene in breeding (Jin, Szabo, Pretorius, Singh, Ward & Beach 2008). Nearly all Kenyan germplasm are known to be susceptible or partially susceptible to *Ug99* (Njau, Keller, Macharia, Singh & Wanyera, 2009). The stem rust resistance gene *Sr36* confers a near-immune resistance reaction to many races of stem rust and is highly effective against race *TTKSK*, which possesses unusually broad virulence combinations. Because this gene is widely used in United States soft winter wheat germplasm and cultivars, it has been considered

to be an important source of resistance to *TTKSK* (Jin, Szabo, Pretorius, Singh, Ward & Fetch, 2008).

The spread of *Ug99* race group of stem rust in eastern and southern Africa and beyond has brought back stem rust research and development activities back onto the international wheat improvement agenda under the BGRI (Borlaug Global Rust Initiative), (Singh, Hodson, Jin, Lagudah, Ayliffe, Bhavani, Rouse, Pretorius, Szabo, Huerta-Espino, Basnet, Lan & Hovmoller, 2015). Currently, the research of stem rust in wheat is focusing on identifying further resistance genes to control *Ug99* and its derivatives (Haile & Roder, 2013). Despite the identification and deployment of a number of rust resistance genes to protect wheat crops, the emergence of virulent pathogen pathotypes can restrict their durability and use (Pathan & Park, 2006). Therefore resistance in wheat varieties has to be constantly improved to avoid having susceptible genotypes in production. Genetic improvement to minimize yield loss under disease is an attractive goal, as it exerts little or no selection pressure on pathogen populations, and could form a useful component of durable disease management programme (Bingham, Walters, Foulkes & Powley, 2009). Because of this, there is a constant need to identify, characterize and deploy new sources of resistance (Pathan & Park, 2006). World population is increasing and food security is projected to become more critical therefore increasing wheat yield potential in the developing world remains a high priority (Duveiller, Singh & Nicolas, 2007). Breeding resistant wheat varieties that have superior yields compared to currently grown popular varieties is a useful option (Singh, Hodson, Huerta-Espino, Jin, Bhavani, Njau, Herrera-Foessel, Singh & Govindan, 2011).

2.2 Materials and Methods

2.2.1 Seedling Stage Experiment

The seedling stage experiment was conducted KALRO (Kenya Agricultural & Livestock Research Organization) - Njoro where all the fifty genotypes were tested for resistance. The infection type at seedling stage was done following the procedures by McIntosh, *et al.* (1995). The seedlings were assessed for disease severity.

3.3.2 Experimental Genotypes

The numbers of genotypes were 45 from the advanced wheat selection group and five local checks of the commonly grown varieties (Appendix 1). The genotypes were made up of bread wheat as described in the Appendix 1. The advanced genotypes were mainly a selection from the CIMMYT (International Maize and Wheat Improvement Center) durable resistance rust nursery. The genotypes were selected continuously over seasons and tested both in Kenya and Mexico, showing promising traits for both yield and stem rust resistance.

3.3.3 Inoculum Preparation for Seedling Stage Resistance

The inoculum used was collected from the trap nurseries of KALRO Njoro in the evening when it was cold. The trap nurseries were planted using the highly susceptible variety *Cacuke* for high amounts of Urediniospores used for inoculation. The trap nurseries were planted early before the main crop. It contained a bulk of Urediniospores of the common two races of *TTKST* and *TTKSK*. The inoculum was made up of a mixture of pathotypes for both *TTKST* and *TTKSK* stem rust races. The inoculum was measured based on the amount of spore number per unit weight using a hemacytometer 1 dilute spores in a 1:1 mixture of Soltrol oil. The solution was placed on a glass slide using a pipette, and loaded on the hemocytometer and placed on the microscope stage, the counting grid was brought to power focus. Spores were counted in selected squares. The spores counted were calculated for concentration using the formula below:

$$\text{Total cells/ml} = \frac{\text{Total cells counted} \times \text{dilution factor} \times 10,000 \text{ cells/ml}}{\# \text{ of Squares}}$$

The number of spores per unit dilute spores in a 1:1 mixture had a concentration of 6×10^6 spores/ml of oil. The mixture was used to spray on the genotypes using a hand sprayer.

3.3.4 Seedling Stage Experiment

The experiment was conducted in the greenhouse at the Kenya Agricultural Livestock and Research Organization (KALRO) Njoro. Fifty pots of 5 cm diameter each filled with a potting media (Hygramix) were used for planting ten seeds of the genotypes. The pots were placed in a plastic tray of ten pots each. The inoculated plants were air dried for half an hour. The pots were

... placed in the growth chamber and removed after ten days for inoculation. The inoculum containing a bulk of the stem rust races *TTKST* and *TTKSK* was sprayed on the genotypes and all checks using a hand sprayer. The pots were then kept in a dark humidity chamber for 48 hrs before taking them to the incubation chamber. In the incubation chamber the pots were left until lesions started forming for data collection. To test for resistance the experiment was repeated five times in the greenhouse and data collected was used to determine which genotypes had resistance.

3.5 Data Collection

Data collection was done fourteen days after inoculation when most of the leaves showed infection. Assessment was done to show which genotypes were consistent for low levels of infection. The genotypes were scored following a scale of 0-4 according to Stakman, *et al.* (1962) as described below. The numbers indicate the infection type while the host response is described as immune to very susceptible as follows; 0=immune, ;=nearly immune, 1=very resistant, 2=moderately resistant, X, Y, Z= heterogenous types, 3=moderately susceptible and 4=susceptible. All data was collected and compared for consistency for the seedling stage resistance (Plate 3.1).

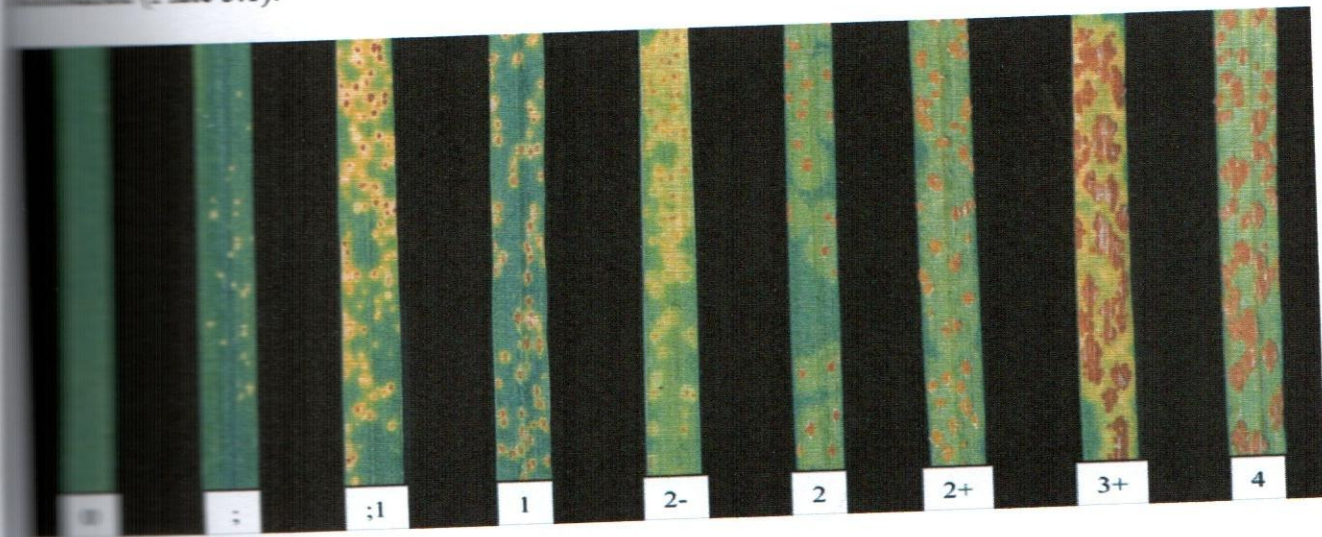


Plate 3.1: Stakman's Infection Type Scale

3.4 Field Experiment

3.4.1 Experimental Locations

The field experiment was conducted at three locations: namely Mau-Narok, Njoro and Lanet. Kenya Agricultural livestock and Research organization (KALRO) field station, situated at Njoro location has an altitude of 2185 meters above sea level (masl), average annual rainfall of 955 mm and minimum and maximum temperatures of 9.7°C and maximum of 23.5°C, respectively. Agricultural Development Corporation (ADC) Enchili farm Mau-Narok is situated at Mau-Narok location has an average annual rainfall of 752 mm, an altitude of 2900 masl and an annual rainfall range of 1,200 to 1,400 mm, minimum and maximum temperatures ranges of 9°C - 14°C and 22°C - 26°C, respectively. Kenya Plant Health Inspectorate Service (KEPHIS) Lanet is situated at Lanet location, 1920 masl with a minimum temperature of 10°C and maximum temperature of 26°C and annual rainfall of 800 mm (Jaetzold, *et al.* 2010).

3.4.2 Experimental Procedure

Land preparation was done by ploughing once and harrowing twice for all the three locations to obtain fine seedbed. The trial design at all the three locations was an alpha lattice of 5 blocks with 10 plots within blocks and replicated three times and plot sizes were 1 m by 2 m. Spacing was 20 cm between rows by drill. Planting was done by hand in all the three locations. The genotypes were tested for resistance to stem rust under natural infection. Genotypes possessing *Sr24* genes with susceptibility to *TTKST* were used as a spreader. Four rows of the *Sr24* susceptible genotypes used as spreader were planted around the experimental plot and between replicates. A seed rate of 125 kg ha⁻¹ which amounts to 25 g plot⁻¹ was used. During planting D.A.P fertilizer was applied at the rate of 22.5 kg of N ha⁻¹ and 25.3 kg P ha⁻¹. At five weeks after planting Urea was used as a nitrogenous fertilizer as a top dress at the rate of 32 kg of N ha⁻¹. Weed control was done using Hussar evolution herbicide at the rate of 0.15 ml 1m⁻². Scoring of stem rust was done when 50% of the susceptible spreader genotypes had been affected. Scoring was done three times across all the locations after twelve days and ten days from the first reading and second reading, respectively.

ACS Data Collection

Data on diseases severity was scored following the modified Cobb scale as described by Peterson, *et al.* (1948). Cobbs scale key of 0.37 representing 1% of the actual affected tissue by disease to 37.0 represented 100% leaves covered by pustules. The percentages indicated the infection type used to determine the disease severity of 0-100%. The host response was assessed as described in Roelfs, *et al.* (1992). The adult plant response to infection in the field was scored using 'R' indicating resistance, 'MR' indicating moderate resistance, 'MS' indicating moderately susceptible, 'S' indicating full susceptibility. The overlapping responses between two categories were indicated using a slash between the two which was MR/MS (Plate 3.2 & 3.3)

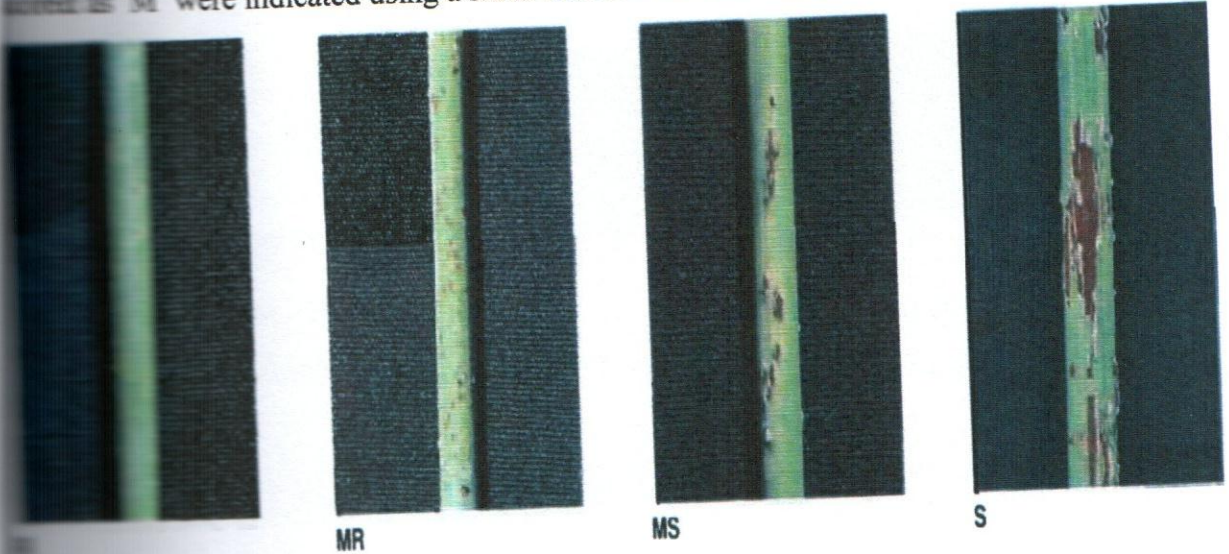


Plate 3.2: Roelfs Field Disease Response to Infection Scale

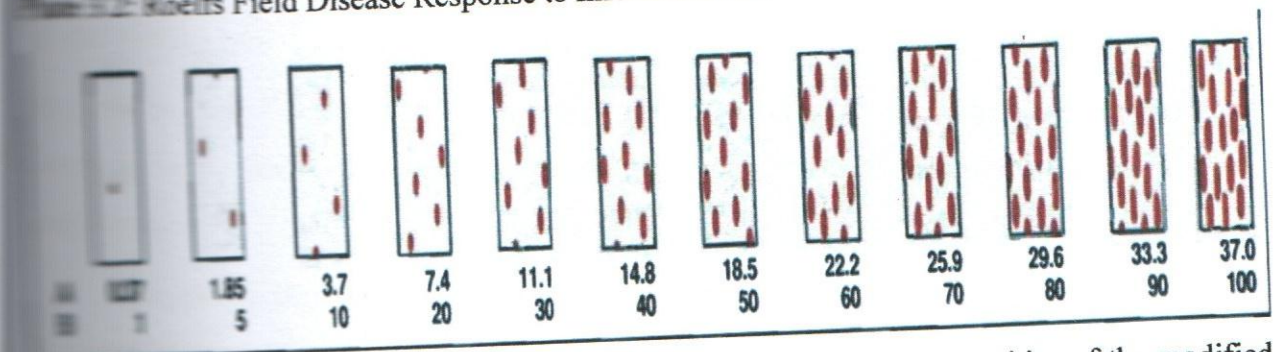


Plate 3.3: Actual percentage occupied by rust Urediniospores; B, rust severities of the modified Cobb scale after Peterson, Campbell & Hannah, (1948).

SAS Yield and Thousand Kernel Weight

Grain yield plot⁻¹ of the entire experimental plots were weighed in grams and converted to tonnes ha⁻¹ for all the plots in the three locations having a total of 450 data entries. The weight of thousand kernels of grains harvested from each experimental plot was also measured. The thousand kernel weight was a yield component.

SAS Data Analyses

The Area Under the Disease Progress Curve (AUDPC) was calculated for all the forty five elite genotypes and five local checks according to the formula from Shaner & Finney, (1977) as given below:

$$\text{AUDPC} = \sum_{i=1}^n [(Y_{i+1} + Y_i)/2] [X_{i+1} - X_i]$$

Y_i = the disease severity at the i^{th} observation, X_i = time in days at the i^{th} observation, n = total number of observations. Analysis of variance was used to find the mean values of AUDPC using SAS version 8.02 (SAS/STAT software 1999). The experimental model was as shown below;

$$Y_{ijkl} = \mu + G_i + R_k + L_j + B_{l(k)} + GL_{ij} + \epsilon_{ijkl}$$

$j=1..3$ $k=1..3$ $i=1..50$ $l=1..5$, Y_{ijk} - overall response of the genotypes

μ - the overall mean, G_i - effect due to the i^{th} genotype in the k^{th} replicate and l^{th} block

$B_{l(k)}$ - effect of the l^{th} block in the k^{th} replicate, R_k - effect due to k^{th} replicate, L_j - effect due to j^{th} location, GL_{ij} - interaction between the i^{th} genotype, j^{th} location and ϵ_{ijkl} - random error component.

Analysis for stability of the genotypes was done using the variance (S_i^2) for a genotype across environments. The S_i^2 was used to determine the most stable genotype on disease across the three locations using the formula described by Francis & Kannenberg (1978) as shown below.

$$S_i^2 = \sum_{i=j}^q (x_{ij} - \bar{x}_i)^2 / q-1,$$

Where S_i^2 is the variance for a genotype across environments, q = number of locations, x_{ij} is the observed mean of the genotype, \bar{x}_i =the mean of the genotype i in the three locations. The Coefficient of Variation of each genotype (CV_i) was used to determine the most stable line on disease and yield across the three locations using formula described by Francis & Kannenberg (1973) as shown below was used for

$$CV_i = S_i / \bar{x}_i \times 100$$

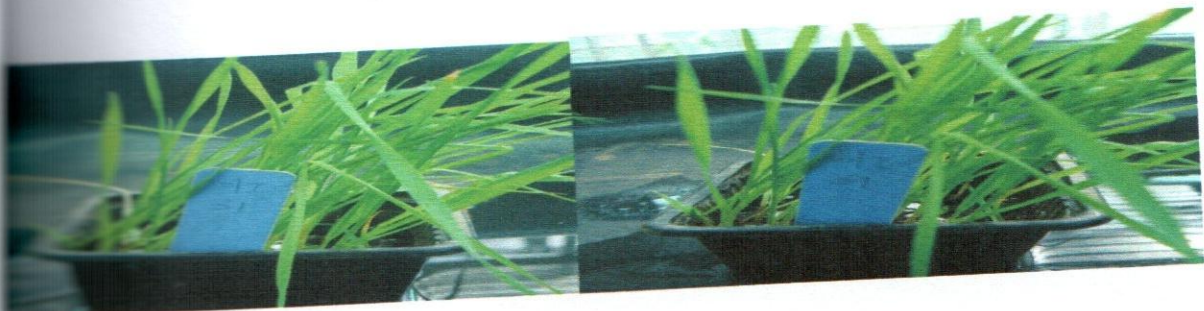
Where CV_i is the coefficient of variation of each genotype in percentage, S_i is the standard deviation for each genotype while \bar{x}_i is the mean of the genotype i across locations.

The correlation coefficient r between yield and AUDPC and between yield and final disease severity was calculated following the formula from Mead, *et al.* (1993).

2.5 Results

2.5.1 Seedling Stage Resistance Experiment

Variation was observed among the genotypes for seedling stage infection after a repeated score of five times (Table 3.1). From the results considering top 24 genotypes in (Table 3.1). The genotypes with small sized Uredinia surrounded by necrosis were very resistant and these were genotype KSL50, 31, 44, 54, 51, 156, 81 and KSL33 (Plate 3.4). The genotypes with medium uredinia often surrounded by chlorosis or necrosis were moderately resistant (Plate 3.5). Genotype KSL144, 115, 146, 69, 76, 161, 53, 137, 37, 52, 17 and KSL 57 with medium uredia were moderately resistant (Plate 3.5). On the other hand genotype KSL 142, 71, 72 and KSL73 with Medium Uredinia and chlorosis were moderately susceptible (Plate 3.6). Genotypes with large uredinia without chlorosis were susceptible (Plate 3.7). The best performing genotypes at seedling stage resistance were entry KSL 144 (2+), 50 (1+), 31 (1+), 44 (1+), 115 (2+), 146 (2+), 156 (2+) and 76 (2+) (Table 3.1). The top performing genotypes accounted for 32% of the total number of genotypes as being very resistant. The moderately resistant genotypes were 44% and 24% were moderately susceptible.



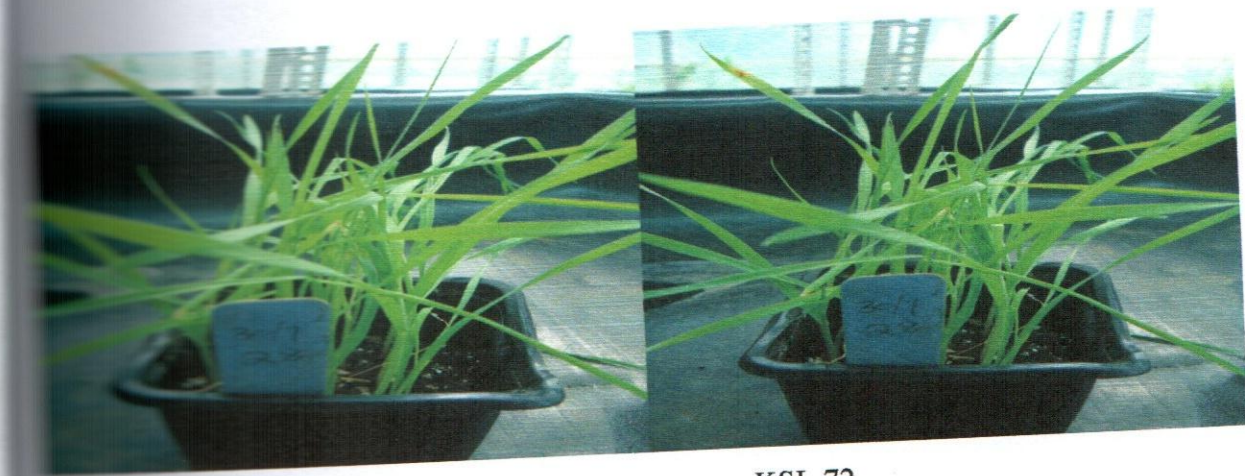
KSL 50

Figure 3.4: Very Resistant genotypes at seedling stage in the green house



KSL 144

Figure 3.5 Moderately resistant genotypes at seedling stage in the green house



KSL 72

Karungo

Figure 3.6: Moderately Susceptible genotypes at seedling stage in the green house



KSL 73

Figure 3.7: Moderately Susceptible genotypes at seedling stage in the green house

Table 3.1: Seedling stage reaction on stem rust based on the AUDPC values from the three locations of Mau-Narok, Njoro and Lanet.

Genotypes	Seedling Infection Types	Host Response
KSL142	3+	Moderately susceptible
KSL71	3+	Moderately susceptible
KSL144	2+	Moderately resistant
KSL50	1; +	Very resistant
KSL31	1; +	Very resistant
KSL44	1+	Very resistant
KSL115	2+	Moderately resistant
KSL146	2+	Moderately resistant
KSL69	2+	Moderately resistant
KSL161	2+	Moderately resistant
KSL53	2+	Moderately resistant
KSL73	3+	Moderately susceptible
KSL54	1+	Very resistant

Table 3.1: Continued

Genotypes	Seedling Infection Types	Host Response
KSL51	1+	Very resistant
KSL156	1+	Very resistant
KSL81	1+	Very resistant
KSL137	2+	Moderately resistant
KSL 37	2+	Moderately resistant
KSL72	3+	Moderately susceptible
KSL52	2+	Moderately resistant
KSL33	1+	Very resistant
KSL17	2+	Moderately resistant
KSL57	2+	Moderately resistant
Checks		
Kingbird ^a	2+	Moderately resistant
Eagle 10 ^a	1+	Very resistant
Korongo ^a	3+	Moderately susceptible
Kenya Wren ^a	3+	Moderately susceptible
Robin ^a	3+	Moderately susceptible

KSL: Kenyan Selection KEY: ;= Near immune 1=Very resistant, 2=Moderately resistant 3=Moderately susceptible 4=Susceptible^a: Local checks

Performance of Genotypes Across Location

The analysis of variance among genotypes for Area Under Disease Progress Curve (AUDPC), Final Disease Severity (FDS), yield and 1000-kernal weight was performed using SAS version 8.02 (SAS/STAT software 1999). The Analysis of variance (ANOVA) for AUDPC revealed variation among the genotypes and locations at $P < 0.05$, $P < 0.01$ and $P < 0.001$ being highly significant ($P < 0.05$) (Appendix 2). The locations, genotype, genotype and location

interaction were highly significant for Final Disease severity (FDS). The genotype and location for grain yield was highly significant. The AUDPC values for the genotypes showed regularity with the FDS values (Table 3.2).

The values for Area Under the Disease Progress Curve (AUDPC) ranged from KSL 142 (209). The diseases severity progressed as the growth of plant increased the first reading had low disease levels by the third reading the levels had increased. Mau-Narok had the highest mean AUDPC at 363.18 followed by Njoro at 326.87 and Lanet at 231.95. The genotypes AUDPC levels were consistent in performance among the locations. Mau-Narok had consistently higher AUDPC values for the genotypes as compared to Njoro and Lanet (Appendix 3).

3.3.3 Stem rust Disease Effect on Genotype Yield and Thousand Kernel Weight (TKW)

There were significant differences in yield among the genotypes. The highest mean yield for the genotypes was at Mau-Narok with 2.82 t ha^{-1} followed by Njoro 1.27 t ha^{-1} and then Lanet 1.25 t ha^{-1} . Performance of genotypes for yield varied from one location to the other. In Mau-Narok the highest grain yield was obtained by KSL 137 (2.63 t ha^{-1}), 31 (2.52 t ha^{-1}), 50 (2.46 t ha^{-1}) and KSL 33 (1.98 t ha^{-1}) (3.3). The same genotypes performed well in Njoro and Lanet. Mau-Narok had better grain yield per genotype with Lanet having less grain yield per genotype. The thousand kernel weight for locations showed Njoro having high quality grains better than Mau-Narok where as Lanet the grains were less quality showed by mean values (Table 3.3). The thousand kernel weight was not significant for the genotypes or location.

Table 3.3: Area Under Disease Progress Curve (AUDPC) and Final Disease Severity means for the best twenty five genotypes in the three locations

Genotype	AUDPC					Final Disease Severity				
	Lanet	Njoro	Mau-Narok	Means	Lanet	Njoro	Mau-Narok	Means		
KSL142	0.0	25.8	60.8	28.9	0.0	0.0	8.3	2.8		
KSL71	75.0	5.8	27.5	36.1	5.0	1.7	3.3	3.3		
KSL144	0.0	25.8	82.5	36.1	0.0	0.0	10.0	3.3		
KSL50	11.7	0.0	110.0	40.6	5.0	0.0	15.0	6.7		
KSL31	60.8	5.8	137.5	68.1	8.3	1.7	16.7	8.9		
KSL44	33.3	31.7	141.7	68.9	5.0	1.7	13.0	6.5		
KSL115	33.3	170.0	72.5	91.9	5.0	0.0	11.7	8.9		
KSL146	76.7	63.3	165.8	101.9	8.3	3.3	11.7	7.8		
KSL69	98.30	112.5	140.0	116.9	11.6	10.0	11.7	11.1		
KSL76	45.00	96.7	211.7	117.8	8.30	8.3	33.3	16.7		
KSL161	88.30	213.3	66.7	122.8	11.7	13.3	10.0	11.7		
KSL53	94.30	152.5	165.0	137.2	13.3	5.0	21.7	13.3		
KSL73	17.50	258.3	151.7	142.5	5.00	20.0	13.3	12.8		
KSL54	110.0	217.5	131.7	153.1	13.3	11.7	15.0	13.3		

Genotype	Lanet	Njoro	Mau-Narok	Means	Lanet	Njoro	Mau-Narok	Means
KSL51	215.0	69.2	180.8	155.0	16.7	5.0	23.3	12.8
KSL156	167.0	130.0	169.0	155.6	16.7	11.7	16.7	15.0
KSL81	141.0	76.7	267.5	161.9	15.0	10.0	30.0	18.3
KSL137	66.70	5.8	438.3	170.3	10.0	1.7	50.0	20.1
KSL	88.30	245.0	204.2	179.2	11.7	15.0	28.3	18.3
KSL72	120.0	221.7	296.5	212.8	13.3	11.7	40.0	21.7
KSL52	157.5	154.2	351.7	221.2	16.7	10.0	43.3	23.3
KSL33	82.50	167.5	416.7	222.2	10.0	16.7	50.0	25.6
KSL17	120.0	171.7	385.0	225.6	13.3	23.3	46.7	27.8
KSL57	100.0	290.8	328.3	239.7	15.0	11.7	40.0	22.2
Checks								
Kingbird ^a	280.0	295.0	177.5	250.8	23.3	8.30	13.0	14.9
Eagle 10 ^a	480.0	398.0	225.0	367.8	33.3	32.3	16.0	27.2
Korongo ^a	698.3	686.7	395.0	593.3	53.3	28.3	53.3	45.0
Kenya Wren ^a	530.0	745.0	623.3	632.8	50.0	53.3	70.0	57.8

Table 3.3: Continued

AUDPC

Final Disease Severity

Genotype	Lanet	Njoro	Mau- Narok	Means	Lanet	Njoro	Mau-Narok	Means
Robin ^a	875.8	970.0	1093.0	979.7	45.0	80.0	80.0	68.3
Means	232.0	326.9	363.2	307.3	23.9	23.3	35.7	27.6
CV%	36.2				CV%	36.2		

LSD 0.05 between locations 25.3

LSD 0.05 between locations 2.09

LSD 0.05 within locations 103.3

LSD 0.05 within locations 8.513

KSL: Kenyan Selection, ^a: Local checks

Table 3.3: Grain yield per plot in t/ha for the three locations and thousand kernel weights of the best performing twenty five genotypes

Genotype	Grain yield in t/ha					Thousand Kernel Weight in grams				
	Lanet	Njoro	Mau-Narok	Means	Lanet	Njoro	Mau-Narok	Means		
KSL 142	0.64	2.19	0.99	1.28	0.03	0.03	0.04	0.03		
KSL 71	0.54	1.01	2.46	1.33	0.03	0.03	0.03	0.03		
KSL 144	0.57	1.65	1.70	1.31	0.02	0.03	0.04	0.03		
KSL 50	0.57	2.19	4.63	2.46	0.03	0.03	0.03	0.03		
KSL 31	0.77	2.18	4.63	2.52	0.03	0.03	0.03	0.03		
KSL 44	0.63	1.93	2.96	1.84	0.03	0.03	0.02	0.03		
KSL 115	0.26	1.43	2.10	1.26	0.03	0.03	0.03	0.03		
KSL 146	0.70	1.70	2.03	1.48	0.02	0.03	0.03	0.03		
KSL 69	0.35	1.20	2.12	1.22	0.02	0.03	0.02	0.02		
KSL 76	0.43	1.96	2.48	1.63	0.02	0.03	0.03	0.03		
KSL 161	0.61	1.82	2.84	1.76	0.03	0.04	0.02	0.03		
KSL 53	0.60	1.38	4.79	2.26	0.02	0.03	0.03	0.03		
KSL 73	0.38	1.57	3.19	1.71	0.02	0.03	0.02	0.02		
KSL 54	0.834	2.01	2.10	1.65	0.02	0.03	0.02	0.03		

Table 3.1 Continued

Grain yield in t/ha

Thousand Kernel Weight in grams

Genotype	Lanet	Njoro	Mau-Narok	Means	Lanet	Njoro	Mau-Narok	Means
KSL 51	0.55	1.86	1.98	1.46	0.02	0.03	0.03	0.03
KSL 156	0.42	1.59	3.32	1.78	0.02	0.03	0.02	0.03
KSL 81	0.23	1.37	1.63	1.08	0.02	0.03	0.03	0.02
KSL 137	0.84	2.01	5.03	2.63	0.03	0.03	0.04	0.02
KSL 37	0.26	1.04	2.75	1.36	0.02	0.03	0.03	0.02
KSL 72	0.31	1.29	3.44	1.68	0.02	0.02	0.03	0.02
KSL 52	0.51	1.61	1.62	1.26	0.02	0.03	0.03	0.02
KSL17	0.60	1.02	3.69	1.77	0.02	0.03	0.02	0.02
KSL33	1.16	1.29	3.82	1.98	0.02	0.03	0.03	0.02
KSL57	0.29	1.39	1.74	1.14	0.02	0.02	0.03	0.02
Checks								
Kingbird ^a	0.48	0.87	3.16	1.51	0.02	0.02	0.03	0.03
Eagle 10 ^a	1.20	1.09	2.40	1.28	0.01	0.03	0.02	0.02
Korongo ^a	0.48	2.18	3.20	1.96	0.02	0.02	0.02	0.02
Kenya Wren ^a	0.49	0.25	2.79	1.45	0.02	0.03	0.03	0.02

Table 3.3. Continued

Robin ^a	1.14	1.09	1.24	1.16	0.02	0.02	0.02	0.02
Means	0.51	1.27	2.82	1.53	0.02	0.03	0.03	0.02
	CV% 54					CV% 34		

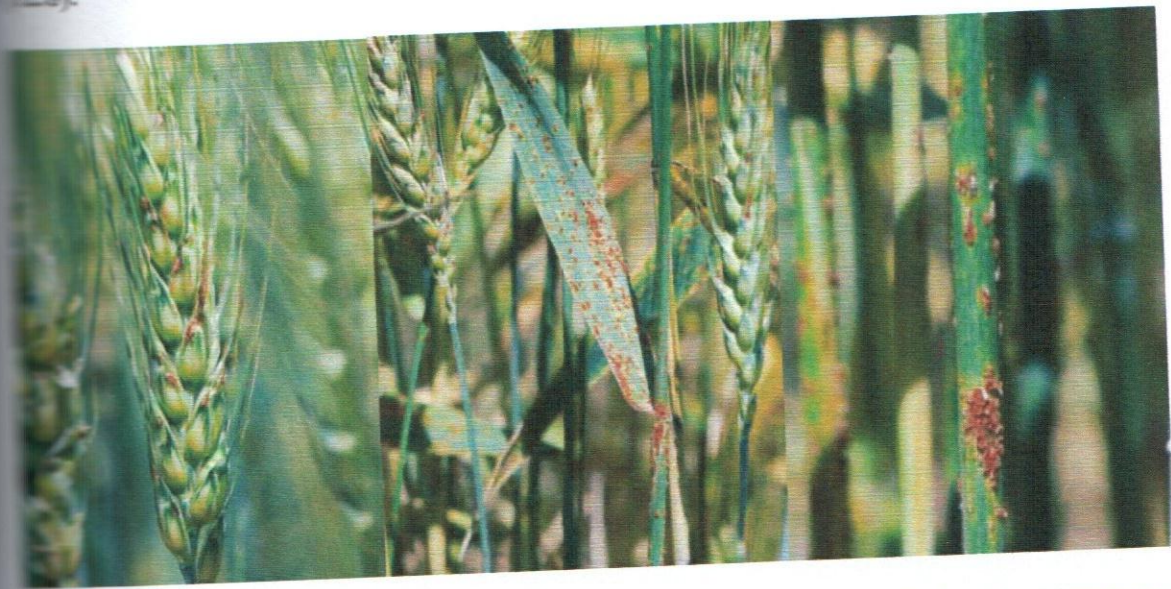
LSD 0.05 between locations 0.188 LSD 0.05 between locations 0.0019

LSD 0.05 within locations 0.769 LSD 0.05 within locations 0.079

KSL: Kenyan Selection, ^a: Local checks

3.4 Adult Plant Response to Infection for Genotypes

In Lanet the genotypes that had a resistant (R) reaction to stem rust were KSL 142, 71 and 144. The ones possessing a moderately resistant (MR) reaction were genotypes KSL 161, 69, 156, 81, 137 and 57. The genotypes with moderately resistant to moderately susceptible (M) were KSL 44, 115, 146, 76, 53, 73, 54, 51, 72, 33 and 17 (3. 4). The genotype with moderately susceptible reaction (MS) was 52. In Njoro the genotypes that had a resistant reaction were KSL 142, 144, 50, 31, 115 and 137. Genotypes possessing moderately resistant reaction were KSL 142, 144, 44, 37 and 57. The genotypes with moderately resistant to moderately susceptible were KSL 69, 76, 53, 54, 51, 156, 72, 33 and 17 across the three locations which also reported high grain yields. The genotypes KSL 52 were moderately susceptible. In Mau-Narok most of the genotypes showed a moderately susceptible reaction which were genotypes KSL 69, 76, 161, 53, 73, 54, 37, 72, 52, 33, 17 and 57. The genotypes with resistant to moderately resistant were KSL 142(2.80), 71 (3.30), 144 (3.30), 31 (6.70), 115 (8.90), 146 (8.90), 156m (6.50) and 137 (8.3). The genotypes with moderately resistant reaction were KSL 50 (6.70), 44 (6.50) and 51 (2.8).



Stem rust on the ear

Leaves bearing spores

Stems of wheat bearing spores

Plate 3.8 Different features of stem rust



Kingbird

KSL 142

Plate 3.8.1: Resistant Local check and genotype



Eagle 10

KSL 44

Plate 3.8.2: Local check Moderately Resistant and genotype



Korongo

KSL 81

Plate 3.8.3: Moderately Susceptible Local check and genotype



Robin

KSL 13

Plate 3.8.4: Susceptible Local check and genotype

Table 3.4: Adult Host response for stem rust infection of the genotypes across the three locations

Genotype	Lanet	Njoro	Mau-Narok
KSL 142	R	MR	M
KSL 71	R	R	M
KSL 144	R	MR	M
KSL 50	MR	R	MR
KSL 31	M	R	M
KSL 44	M	MR	MR
KSL 115	M	R	M
KSL 146	M	M	M
KSL 69	MR	M	MS
KSL 76	M	M	MS
KSL 161	MR	MR	MS
KSL 54	M	M	MS
KSL 51	M	M	MR
KSL 156	MR	M	M
KSL 81	MR	MR	MS

Table 3.4: Continued

Genotype	Lanet	Njoro	Mau-Narok
KSL 37	M	MR	MS
KSL 72	M	M	MS
KSL 52	MS	MS	MS
KSL 33	M	M	MS
KSL 17	M	M	MS
KSL 57	MR	MR	MS
Checks			
Kingbird	MR	MR	M
Korongo	M	MS	MSS
Eagle 10	M	M	MS
Kenya Wren	MS	M	MS
Robin	MSS	MSS	S

R-Resistant, MR- Moderately Resistant, M- Moderately Resistant to Moderately Susceptible, MS-Moderately Susceptible, MSS- Moderately susceptible to Susceptible, S-Susceptible

R=0-15% Severity, MR=15-30% Severity, M=20-25% Severity, MS=30-40% Severity

MSS=50-60% Severity S=60-100% severity.

3.5.5 Genotypic Stability

The coefficient of variation (CV_i) and variance (S^2_i) identified stable genotypes across the three locations. Generally, stable genotypes had lower values of CV_i and S^2_i compared to those that were less stable (Table 3.5). Amongst the genotypes, the most stable were KSL 69, 161, 54 and 156 with less than 20% coefficient of variation values. While the most unstable had higher values which were KSL 137, 44 and 76 among the top twenty four. Genotype KSL 21, 58, 42 and 16 were the least stable. The values were directly proportional to each other, when the variance increased the coefficient of variation also increased. The yield data (Table 3.5) showed that the genotypes were very unstable, the CV_i percentage ranged from 42.93% to 98.8% which

were far from the acceptable 20% (Table 3.5). The most stable genotype had 0-20% CV_i and least unstable had 20% and above.

Table 3.5: Coefficient of variation (CV_i) and variance (S_i²) for the top twenty four genotypes based on the FDS values and yield

Genotype	FDS S _i ²	FDS CV _i	Yield S _i ²	Yield CV _i
KSL142	3.6	31.1	0.7	63.9
KSL71	2.7	49.5	1.0	74.9
KSL 144	8.3	43.5	0.4	48.9
KSL50	39.7	81.1	4.2	82.8
KSL31	56.5	84.2	3.8	77.2
KSL44	35.7	89.6	1.4	63.6
KSL115	12.1	38.2	0.9	74.1
KSL146	17.3	55.0	0.5	46.8
KSL69	1.0	8.8	0.8	72.2
KSL76	108.3	86.6	1.1	65.5
KSL161	2.9	14.9	1.3	63.8
KSL53	69.7	62.6	5.0	98.9
KSL73	56.5	58.7	2.0	82.8
KSL54	2.7	12.4	0.1	42.9
KSL51	85.9	62.0	0.7	54.2

Table 3.5: Continued

Genotype	FDS	S_i^2	FDS CV_i	Yield S_i^2	Yield CV_i
KSL81	108.3		56.7	0.6	69.4
KSL156	7.3		18.2	2.1	82.1
KSL137	433.3		96.7	4.7	82.2
KSL37	166.5		70.4	0.9	74.1
KSL72	252.7		73.4	2.6	95.2
KSL52	310.2		75.4	0.4	50.9
KSL33	458.9		83.7	2.6	80.8
KSL17	371.9		78.9	2.8	98.8
KSL57	239.5		69.7	0.6	66.4
Checks					
Kingbird ^a	58.3		50.0	2.7	96.2
Eagle10 ^a	91.9		34.5	1.2	84.3
Korongoa	203.3		32.0	1.9	70.3
Kenya Wren ^a	114.9		18.5	1.4	82.3
Robin ^a	408.3		29.6	1.4	6.7

KSL; Kenyan selection, FDS: Final Disease Severity, S_i^2 : Variance, CV_i : Coefficient of Variation, KSL: Kenyan Selection, ^a Local checks

3.5.6 Correlation Between Yield, AUDPC and Final Disease Severity

The correlation coefficient (r) for AUDPC and grain yield was found to be - 0.943, while coefficient of determination (r^2) was 0.890 (Figure 3.1). Similarly Final Disease Severity and yield r was -0.84 and r^2 was 0.705 (Figure 3.1 and 3.2). The r value revealed a strong negative

relationship between yield and AUDPC and also for yield and FDS explaining 89% of the variation. For the yield and FDS relationship 70.5% was explained.

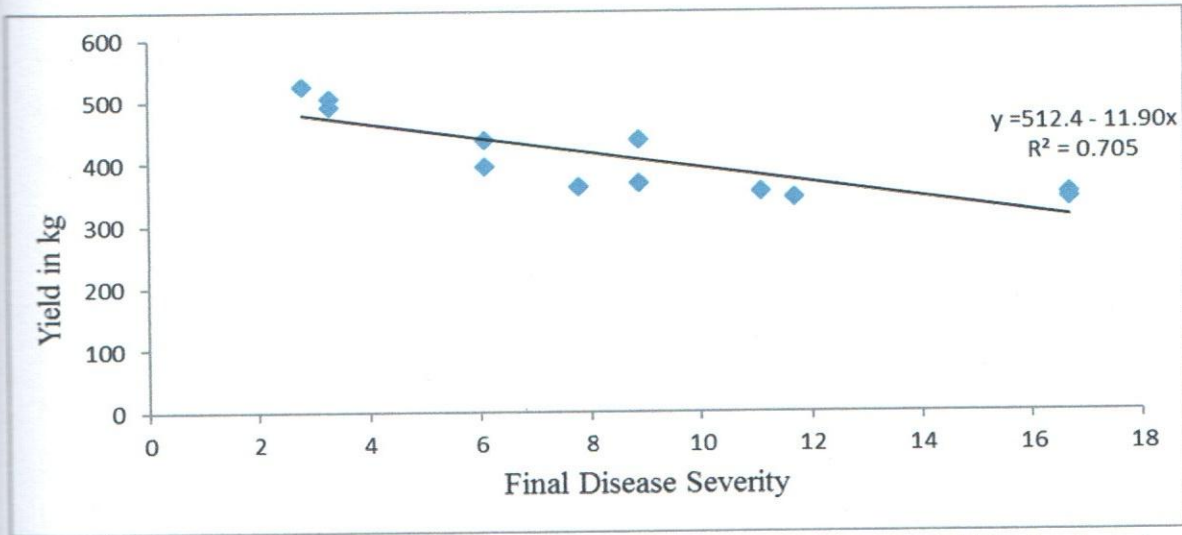


Figure 3.1: Relationship between Final Disease Severity and genotype yield in the three locations of Mau-Narok, Njoro and Lanet

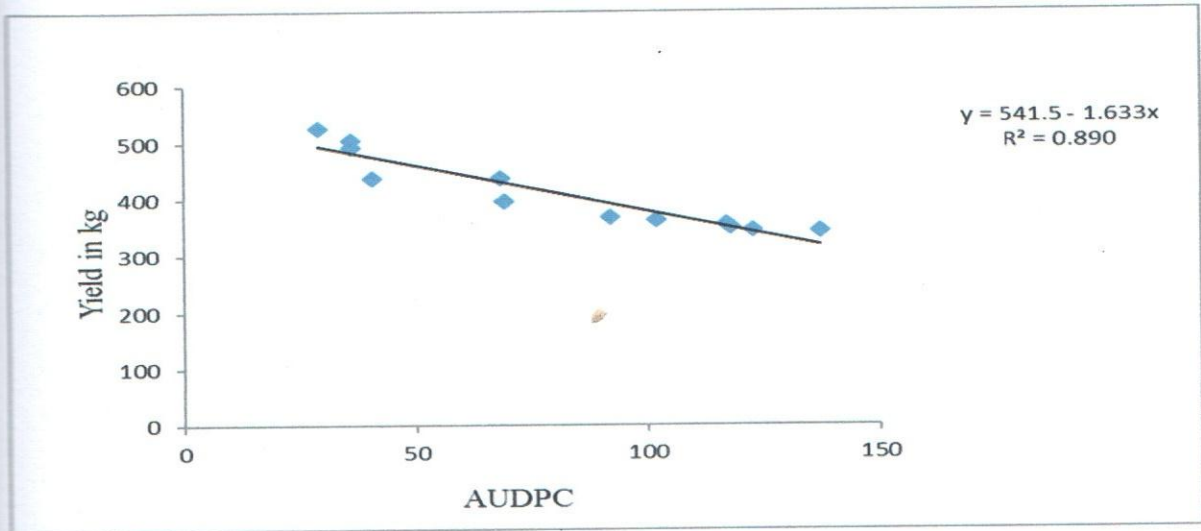


Figure 3.2: Relationship between AUDPC and genotype yield in the three locations of Mau-Narok, Njoro and Lanet

3.6 Discussion

3.6.1 Seedling Stage Resistance

In the seedling stage resistance 42% of the genotypes evaluated had adequate resistance levels of 1+ and 2+ for infection types and being very resistant and moderately resistant

respectively for host response. The remaining 58% had inadequate resistance of expressed susceptible. Seedling resistance according to Pathan & Park (2006) by comparison, is effective at all growth stages. As suggested by GRDC, (2012) protection at the seedling stage is provided by 'major' or seedling resistance genes, which have much larger effect and often provide complete resistance at all growth stages.

3.6.2 AUDPC, FDS, TKW and Yield

ANOVA for the four parameters showed that there was a highly significant genotype and location interaction for FDS and AUDPC ($P < 0.001$), for yield it was only significant at $P < 0.05$. Finlay & Wilkinson, (1963) illustrated that genotypic adaptability has proved to be of particular importance, because edaphic variation between localities and the seasonal variation in any one locality are very great. Thus the mean values for Mau-Narok were slightly high for AUDPC at 363.18 much higher than Lanet but comparable to Njoro at 231.97 and 326.57 respectively. Genotype KSL 142, 71, 144, 50, 31 and 44 showed resistance to stem rust disease across the three locations. At Mau-Narok all the genotypes had high disease severity levels.

Performance among the genotype for grain yield was varied across the locations. The top performing genotypes for grain yield were KSL 137 (2.63 t ha^{-1}), KSL 31 (2.52 t ha^{-1}), KSL 50 (2.46 t ha^{-1}) and KSL 53 (2.63 t ha^{-1}) as the best performing across the three locations. The TKW showed less variation among the genotypes except for location which was not significantly different ($P < 0.05$). As stated by Iftikhar, *et al.* (2012) the thousand kernel weight has a positive direct effect on yield and may also be used as a selection criterion for superior genotypes. Mohammadi, *et al.* (2012) established that grain yield in wheat is frequently the sink limited, and for this reason, the 1000 kernel weight has been reported as a promising trait for increasing grain yield in wheat under different conditions.

3.6.3 Genotype by Environment (Location) Interaction

There were variations in genotypic performance among the three locations in Lanet the genotypes KSL 137, 54, 31, 146, 44, 161, 17 and KSL 53 had good grain yield performance in Lanet. Genotypes KSL 142, 50, 31, 54, 137, 76, 44, 51, 161 and 146 performed well in Njoro. Genotype KSL 137, 50, 31, 44, 53, 33, 17, 156, 72 and 161 were the best performing in Mau-Narok. As stated by Yan (2002) that the measured yield of each cultivar in each test environment is a mixture of environment main effect (E) genotype main effect (G) and genotype and

environment (GE). According to Yan (2002) that typically E explains most (up to 80% or higher) of the total yield variation and G and GE are usually small. The environments showed that wheat grain yield was significantly affected by environment. As in the case of Mau-Narok reporting greater grain yields confirmed by Kaya & Akcura, (2014) that Grain yield and quality traits were affected more intensely by the E than by the G. Mohamed (2013) added that the large yield variation explained by environments indicated that the environments were diverse, with large differences between environmental means contributing most of the variation in grain yield. The environmental conditions include temperatures and rainfall. The rainfall experienced in Mau-Narok was 1249 mm in the growing season of 2014, as compared to Njoro having 804.5 mm and Lanet 950 mm. The maximum temperatures experienced in Mau-Narok were 22 °C, Njoro having 23.5 °C and Lanet having 22 °C.

3.6.4 Seedling and Adult Stage Resistance of Genotypes

Seedling and adult stage resistance genes as explained by Lagudah (2010) in wheat fall under two broad categories and are referred to as seedling and adult plant resistance (APR) genes. Seedling resistance genes are detected during both the seedling and adult plant stages and as such constitute an all stage resistance phenotype. APR is commonly detected at the post-seedling stage and often as field resistance. Therefore the genotypes that had seedling stage reflected well with resistance in the field. The genotypes that possessed both seedling and adult stage resistance were KSL 144 (2+), 50 (1+), 31 (1+), 44 (1+), 115 (2+), 146 (2+), 69 (2+) and 76 (2+) based on the AUDPC and Final Disease Severity values. According to Wang, *et al.* (2005) all genotypes with APR showed lower values for AUDPC than susceptible cultivars. Apparently most of the best performing genotypes were pedigrees of already released varieties such as Kenya Nyangumi, Kongoni, Kwale, Zabadi, Mbuni, Paka and NjoroBWII. There is therefore need to improve on already released varieties for trends have shown that the agronomic performance is superior. Wang, *et al.* (2005) explained that the adult plant resistance (APR) is of major importance in breeding for an efficient genetic control strategy and added that it is possible to combine major resistance genes and APR genes to achieve durable resistance.

3.6.5 Adult Plant Host Response of Genotypes to Stem rust

Adult plant resistance as acknowledged by Draz, *et al.* (2015) is the most economic and effective means of reducing yield losses caused by the stem rust disease. However, breeding genotypes for disease resistance is a continuous process and new effective sources of resistance need to be added to breeding materials. Through the study Lanet had 12.5% of the genotypes with resistance to stem rust, 29.2% were moderately resistant, 54.2% were between being moderately resistant and moderately susceptible and 4% had a moderately susceptible reaction. In Njoro the genotypes with resistance were 20.8%, moderately resistant 33.3%, moderately resistant to moderately susceptible 41.7% and moderately susceptible 4.2%. In Mau-Narok there were no genotypes showing resistance, 12.5% showed a moderately resistant reaction, 33.3% had moderately resistant to moderately susceptible and 54.2% had moderately susceptibility. The implication of host response across the locations is that there were less than 15% of the genotypes with resistance. There was a tendency where genotypes with resistance (R) or moderately resistance (MR) having good grain yield performance as confirmed by Singh, *et al.* (2015) host response of R, R-MR, MR-MS are better candidates for high yielding potential.

3.6.6 The Relationship Between FDS and Genotype Yield

There was heavy disease pressure evidenced by 90 % FDS values on the spreader rows and genotype Robin especially in Mau-Narok Singh, *et al.* (2008) reported the same in Kenya. The spreader rows of *Sr 24* susceptible genotypes had the highest Final Disease Severity of 90% which implies that the races were mainly *TTKST* and *TTKSK*. The genotype interacted well with the environment. In Njoro genotypes KSL 142, 50, 31, 54, 137, 44, 51 and KSL 146 reported good grain yield ranging from 2.19 t ha⁻¹ to 1.70 t ha⁻¹ with FDS values ranging from 0% - 5%. The genotypes with low FDS values reported high grain yields.

3.6.7 Correlation Coefficient (r) and Coefficient of Determination (r²) for AUDPC and FDS

In the study stem rust severity and yield relationship was explained by the negative and high correlation coefficient ($r=-0.943$) for AUDPC and yield (Figure 3.1). The Final disease Severity and yield was at ($r=-0.839$) (Figure 3.2) also having a strong negative relationship, Jeger (2004) explained that even where disease resistance is a major target in breeding programmes, the effect on yield and productivity is an important trait, thus the additional value of the

relationship between AUDPC and yield components. There is strong evidence from the study that grain yield loss and stem rust disease were highly associated. The coefficient of determination (r^2) was based on the amount of variability in one variable (yield) that was explained by the linear function of the other variable (AUDPC). The same case applied to FDS and yield by Gomez & Gomez, (1984). The correlation values for AUDPC and Final Disease Severity signify that yield losses increased under disease presence in a progressive manner.

3.6.8 Coefficient of Variation (CV_i) and Variance (S_i) Yield and Final Disease Severity

The coefficient of variation (CV_i) was used to determine stability for FDS and yield among the genotypes. According to Yan (2002) visualization of the genotype stability is always an important issue in cultivar evaluation. For FDS KSL 69 (8.8%) 54 (12.38%), 161 (14.9%) and 156 (18.24%) were identified as the most stable with less than 20 % CV_i from Lin, *et al.* (1986) and the most unstable were KSL 137 (96.7%) 44 (89%) and KSL 76 (86.57%) among the top twenty four genotypes. While using the yield data to identify stability most of the genotypes were unstable.

3.7 Conclusion

Both seedling and adult stage resistance were determined for fifty genotypes. The best overall genotypes for both adult and seedling stage resistance were KSL 137, 72, 73, 69, 161, 54 and KSL 156. Consistent in performance for the seedling and adult stage resistance, yield, FDS and thousand kernel weight performances as the best. The same genotypes expressed resistance or moderately resistance host response therefore superior on grain yield. The genotypes should be recommended for production or used for improving the already existing varieties.

CHAPTER FOUR

4.0 WHEAT STEM RUST DISEASE INCIDENCE AND SEVERITY ASSOCIATED WITH FARMING PRACTICES IN THE CENTRAL RIFT VALLEY OF KENYA

4.1 Abstract

Stem rust (*Puccinia graminis* f.sp.*tritici*) is a major disease of wheat (*Triticum aestivum* L.) that occurs more in the main wheat growing regions of Kenya. The objective of the research was to assess the incidence and severity of wheat stem rust during the 2015 growing season. A survey was conducted in Mau-Narok, Njoro and Kabatini regions. During the survey 149 small scale wheat growers' fields were assessed. The results revealed that stem rust incidence for the three study areas ranged from 11.3 to 77.8% and severity 20 to 60%. The survey showed that the incidence and severity were associated with the farming practices such as chemical control, varieties grown, use of certified or uncertified seed and cropping systems. High to moderate incidence and severity levels were found on fields with single spray of a fungicide. Fungicide use varied among growers for stem rust control reported 43.2% Mau-Narok, 38.9% Kabatini and 17.8% Njoro applying fungicide. The varieties grown had a relationship to disease incidence and severity percent levels. The use of uncertified seed by farmers contributed to high disease incidence. About 50.6% growers preferred old varieties mainly Robin and NjoroBWII. Crop rotation was practiced by 97.8% of the farmers being wheat with legumes. A multi-tactic disease management approach mainly optimal fungicide use at recommended rates, planting of certified seed of resistant varieties and crop rotation of legumes with wheat are required as stem rust effective management strategies.

4.2 Introduction

Wheat (*Triticum aestivum* L.) is one of the worlds' most productive and important crops in the 21st century (Curtis & Halford, 2014). It is one of the key staple crops for global food security, providing more than 35 % of the cereal calorie intake in the developing world, 74 % in the developed world and 41 % globally from direct consumption (Shiferaw, Smale, Braun, Duveiller, Reynolds & Muricho, 2013). Due to increased consumption and demand for grain, for

food (Curtis & Halford, 2014) wheat yields must be increased as this is seen as an important strategy to prevent food shortages (Curtis & Halford, 2014).

Wheat is the second most important cereal staple food after maize in Kenya (USAID, 2010). In Kenya it is mostly grown in the Rift Valley, some areas of upper Central region (Nyandarua, Nyeri) and parts of Meru (Timau) (USAID, 2010). In spite of the importance of wheat, plant disease is still a major constraint to its production. Plant diseases have been reported to reduce crop yields worldwide, leading to significant crop losses (Khoury & Makkouk, 2010). Stem or black rust, caused by *Puccinia graminis*, has historically caused severe losses to wheat production worldwide (Njau, Keller, Macharia, Singh & Wanyera, 2009). In most wheat-growing regions of the world, existing environmental conditions will favour stem rust infection, which at times leads to epidemic build-up (Singh, Hodson, Huerta- Espino, Jin, Bhavani, Njau, Herrera-Foessel & Govindan, 2011). The situation is worsened by the fact that susceptible wheat varieties are grown over large areas and that a large proportion of current breeding materials are susceptible to stem rust race *Ug99* and other newly identified races. It implies therefore that the stem rust pathogens have the potential to cause a wheat production disaster that would sourly affect food security (Singh, Hodson, Huerta- Espino, Jin, Bhavani, Njau, Herrera-Foessel & Govindan, 2011). Disease assessment is an essential task in the study of plant disease epidemics and vital to the knowledge of whether disease management practices are successful (Campbell & Neher, 1994). Disease severity evaluation is an important decision support for adoption of strategies and tactics for disease control. The most commonly used method to assess disease severity is visual, (Bade & Carmona, 2011). Disease severity is determined by a function of the degree of infection, colonization, and damage of host tissues. Besides the amount of host development and growth is a function of disease severity (Gaunt, 1995).

Disease incidence is defined as the proportion (0 to 1) or percentage (0 to 100) of diseased entities within a sampling unit. This alternative definition provides the needed generality so that incidence is the proportion (or percentage) of diseased leaves on a plant (Seem, 1984). Incidence and severity are measurements of the same group of entities within a sampling unit. The sampling unit for incidence should be the sampling unit for severity (Seem, 1984).

Integrated disease management (IDM), which combines biological, cultural, physical and chemical control strategies in a holistic way of disease control as opposed to using a single component strategy is a better option apart from being sustainable (Khoury & Makkouk, 2010).

It can be defined as a decision-based process involving coordinated use of multiple tactics for optimizing the control of the pathogen ecologically and economically (Khokhar & Gupta 2014). In practice and in most cropping systems today, emphasis is still being placed on a single technology (Khoury & Makkouk, 2010). Many problems have been associated with fungicide use such as the frequent emergence of fungicide resistance in pathogens and the harmful effects of fungicides to human health and the environment (Khoury & Makkouk, 2010).

Wheat production in Kenya mainly takes place on large- and medium-scale farms, using capital intensive technology. The technology on the medium- and large-scale farms is the same as that in Western Europe (FAO, 2013). In contrast small scale farms operations are smaller as compared to the large and medium (FAO, 2013). The small scale wheat farmers complain of prohibitive production expenses and low production (caused by use of non-certified seeds and low use of inputs) and sub-division of land as a major problem (MOA, 2013). Most large scale farmers are still holding stakes of wheat (FAO, 2013). The cost of key inputs such as seed, pesticides is high for resource-poor farmers. Such high costs lead to low application and adulteration of inputs (GOK, 2010).

4.3 Materials and Methods

4.3.1 Sampled Regions

A survey was conducted in three regions of Nakuru county; Njoro, Mau-Narok and Kabatini regions which represented 25.7 %, 35.1 % and 39.2 % of the study area, respectively (Figure 3). The fields surveyed were planted by the farmers early during the 2015 season. In Mau-Narok there were two major cropping seasons, early and late while Njoro and Kabatini had early. Only the fields with the early crop were surveyed in Mau-Narok. Most small scale growers had planted early in Mau-Narok while most medium and large scale farmers planted late.

The Locations surveyed in Mau-Narok regions were Sururu, Mwisho Wa Lami, Likia and Mau-Narok. Mau-Narok had an average annual rainfall of 752 mm, an altitude of 2900 meters above sea level (masl) and an average annual 1300 mm and temperatures range of 14°C and 26°C, respectively. The second regions surveyed were Njoro which had five locations mainly Piave, lower Piave, Njoro and Kerima. Njoro regions had an altitude of 2185 masl, average annual rainfall of 935 mm and minimum and maximum temperatures of 9.7°C and 23.5°C, respectively. The third regions surveyed were Kabatini with four locations mainly Karunga, Ngecha, Thayu

and Ruguru which had many wheat growers. Kabatini with an altitude at 2135 masl with a minimum temperature of 10°C and maximum temperature of 26°C and annual rainfall of 800 mm (Jaetzold, *et al.* 2010).

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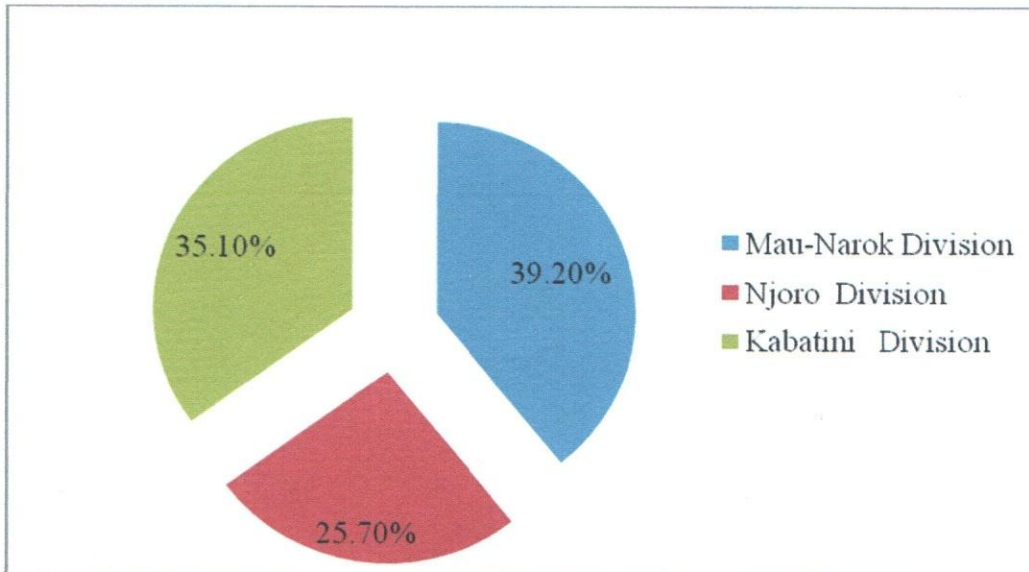


Figure 4.1: The proportion of fields surveyed in the three regions of three regions of Nakuru County, Kenya in 2015

4.3.2 Field Survey

A questionnaire check list was used during the study (Appendix 4). The part one of the questionnaire was about the general information, the name of the division, location and farm classification. Farm classifications were mainly three; small (<10 ha), medium (10-60 ha) or large scale (>60 ha) farms, adapted from MOA (2006). The second part of the questionnaire was about the farming practices in place, fungicide used, rate of fungicide spray used and number of sprays. The other questions were about the wheat varieties commonly grown (Appendix 4).

4.3.3 Sample Size for Disease Assessment

A multi stage sampling technique was applied where fields were grouped as small, medium and large scale in the sampled regions. The sample size of growers selected was done following the formula from Krejcie and Morgan (1970) as shown below;

$$S = \frac{\chi^2 NP(1 - P)}{d^2(N - 1) + \chi^2 P(1 - P)}$$

S = required sample size, χ^2 = the table value of chi-square for 1 degree of freedom at the desired confidence level (3.841), N = the population size, P = the population proportion (assumed to be 0.50 since this would provide the maximum sample size), d = the degree of accuracy expressed as a proportion (0.05). The total sample size was 58 in Mau-Narok, 38 in Njoro and 52 in Kabatini regions.

4.3.4 Assessment of Disease Intensity

A quadrat of 1m by 1m was used for both disease incidence and severity on the same field and 1m² used to obtain the two disease values. The incidence was the number of plant infected by disease and severity was the percentage of foliage attacked by disease on the same plant. The stage of the wheat crop assessed was at the growth stage as stated, Zadoks GS 73 (early milk), GS 75 (medium milk), GS 77 (late milk), GS 83 (early dough) and GS 85 (soft dough) (Zadoks, *et al.* 1974) which was wide-ranging from field to field across the study areas.

A quadrat was cast in the field randomly for the total number of farms visited. The proportion of stem rust infected plants to the total number of plants in the quadrat was calculated from the FAO-SEC, (2012) formula as shown below;

$$DI = \frac{\text{Number of diseased plants in the quadrat}}{\text{Total number of plants in the quadrat}} * 100$$

The same fields and plants used for disease incidence determination were scored for disease severity. Scoring was done following the modified Cobb scale as described by Peterson Campbell & Hannah (1948). The Cobbs scale key of 0.37 representing 1% of the actual affected tissue by disease to 37.0 which represented 100% leaves covered by pustules of 0-100% which was used to determine the disease severity.

4.3.6 Data Analysis

Analysis of data was done using SPSS Version 20 (IBM SPSS Statistics). Data was input for analysis using the descriptive statistics, frequencies and cross tabulation. The frequencies

were in percentages for all the entities. The entities included regions, locations, farm classification, fungicides used, the rates and number of sprays. The percentage for varieties grown, fertilizer use, seed source and a yes and no response for wheat grain yield as being high, medium and low was done. Each component was worked out in percentages among the three regions and arranged in tables accordingly. Disease incidence was calculated directly as a percentage as the number of plant infected over the total number of plants in the quadrat and severity data scored as the percentage of foliage damaged by stem rust disease.

4.4 Results

4.4.1 Stem rust Disease Incidence

Mau-Narok division had 7.9 to 77.8% disease incidence. In Njoro division all the locations surveyed had no stem rust disease incidences. At Kabatini stem rust disease incidence occurred ranged from in 3.3 to 32.1%%. The average disease incidence in Mau-Narok was 32.1 %, Kabatini (7.9%) and Njoro (0%). The absence of disease in Njoro (Figure 4.2) was explained by the growers as an escape due to the changing rainfall patterns. The tendency showed Mau-Narok as having high levels of disease incidence, Kabatini having moderate to low and Njoro no incidence reported.

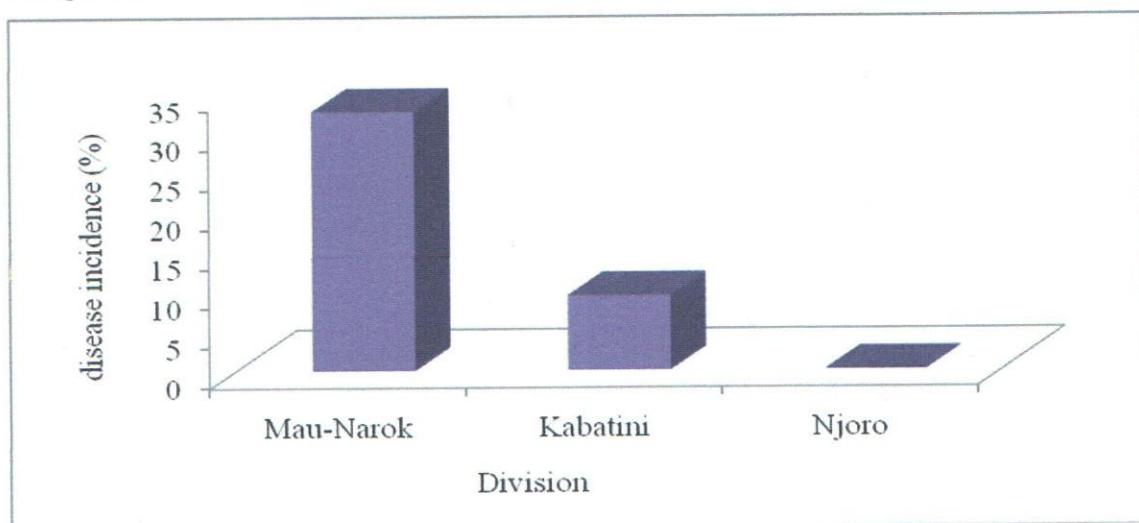


Figure 4.2: The average incidence (%) of wheat stem rust in the three regions of Nakuru County, Kenya in 2015

4.4.2 Stem rust Disease Severity

In Mau-Narok division stem rust disease occurred in severity levels ranging from 30 to 60%. The disease severity in Mau-Narok was high by the figures obtained from all location. Kabatini reported stem rust disease severity range of 20 to 30%. The figures in Kabatini showed that the disease severity levels were low. In Njoro division all the locations surveyed had no stem rust disease severity. The average disease severity in the three regions was Mau-Narok 41.4%, Kabatini 23.3% and Njoro 0% (Figure 4.3).

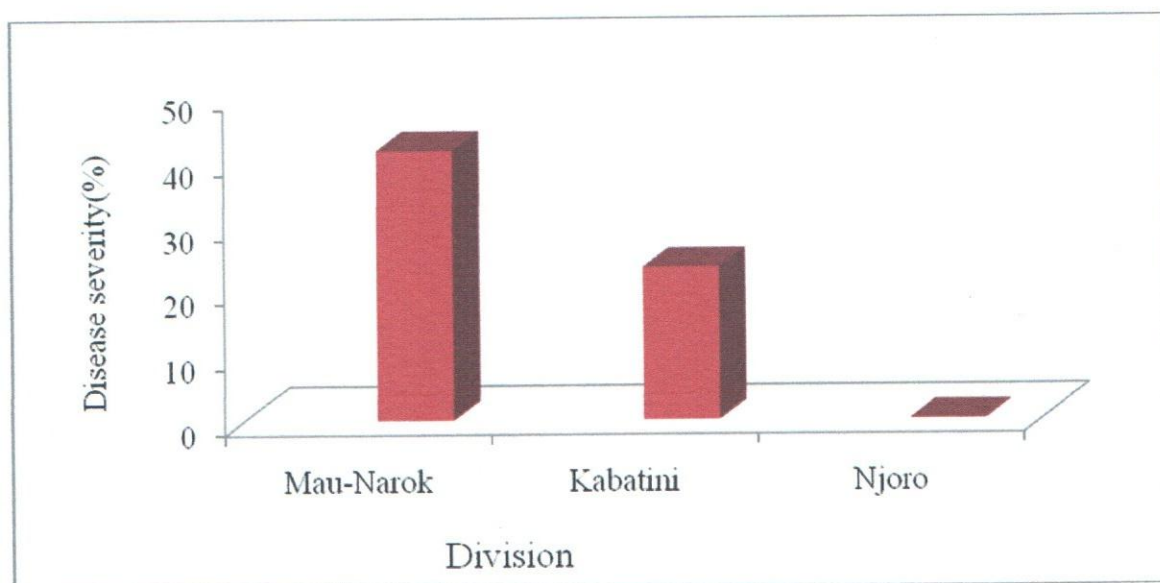


Figure 4.3: The average wheat stem rust severity (%) in three regions of Nakuru County, Kenya in 2015

The disease escape in Njoro was due to the high annual rainfall experienced in 2015 of 951.6 mm during the growing season. The previous season of 2014 had 804.5 mm annual rainfall where stem rust was reportedly high. The low temperatures of 10.5 °C in 2015 as minimum contrasted by a minimum of 11.3 °C in 2014 contributed to disease escape because of unfavorable conditions.

4.4.3 Effect of Fungicide Use, Rate and Number of Sprays on Incidence of Wheat Stem rust

The number of sprays per growing season in Mau-Narok was at 34.5% of the growers spraying once, 34.5% of the farmers sprayed twice while 17.2% of the farmers sprayed thrice.

Fungicide spraying rates at the above recommended in Mau-Narok was 25.9%, at recommended was 60.3% and 13.8% for no spray (Figures 4.4 and 4.5). The farmers were using fungicides rates at above the recommended ones by the manufactures prescriptions. About 42.1% of growers in Njoro did not spray a fungicide on the wheat fields. Few growers (5.3%) used the above recommended rates, recommended rates were 52.6% due to disease escape. In Njoro few growers sprayed their fields once (28.9%). About 26.3% sprayed twice and 2.6% sprayed thrice. The frequency of farmers who did not spray their field stood at 42.1% (Figures 4.4 and 4.5). In Kabatini 9.4% used the above recommended rates, the recommended rates were at 79.2% and those who did not spray at 11.3%. In Kabatini 24.5% sprayed once, 47.1% twice, 15.1% sprayed thrice and 11.3% no spray of fungicide was done (Figures 4.4 and 4.5).

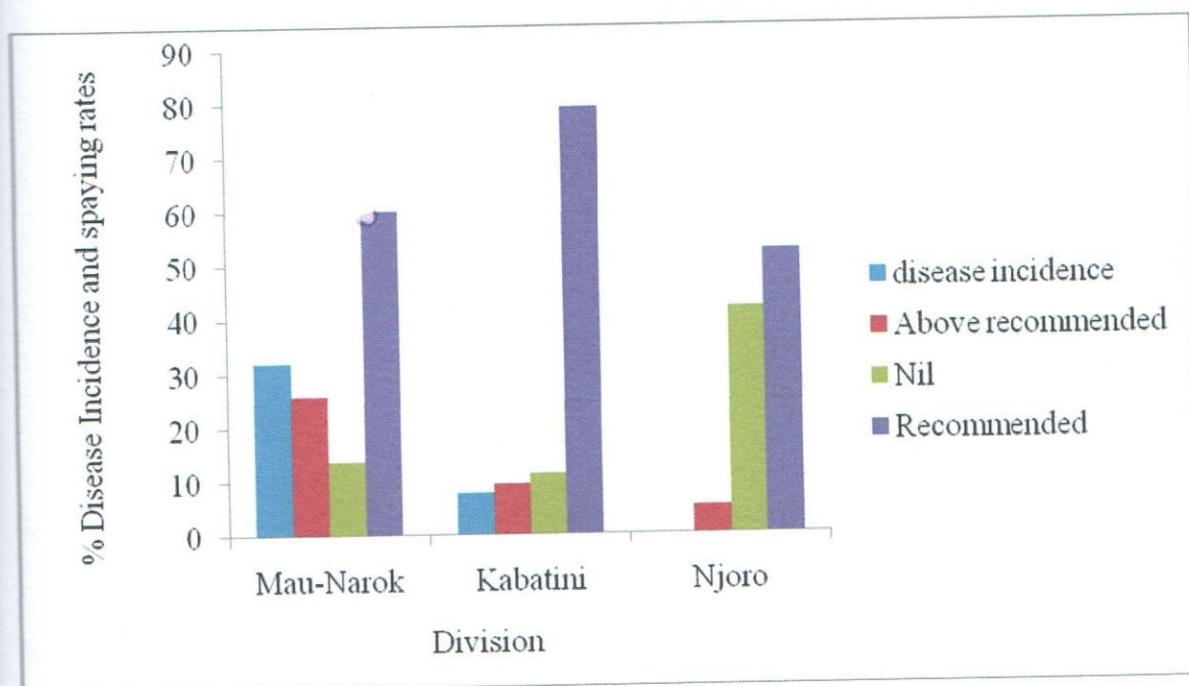


Figure 4.4: Wheat stem rust disease incidence (%) and fungicide spraying rates

A higher percentage of growers using fungicides were found in Mau-Narok, followed by Kabatini and Njoro. Mau-Narok had a large percentage (25.9%) of fields sprayed at the recommended rates. As compared to Njoro and Kabatini, Mau-Narok had the highest percentage of growers of the above the recommended rates 23.9%, respectively. Njoro division had a great number of fields that were not sprayed at 42.1% as compared to Mau-Narok 13.8% and Kabatini

11.3%. Despite the use of above the recommended rates by growers in Mau-Narok disease incidence was moderate to high (Figure 4.2). In Njoro during the growing season there was no disease and growers sprayed only once or did not spray. In Kabatini most of the growers sprayed twice at the recommended rates.

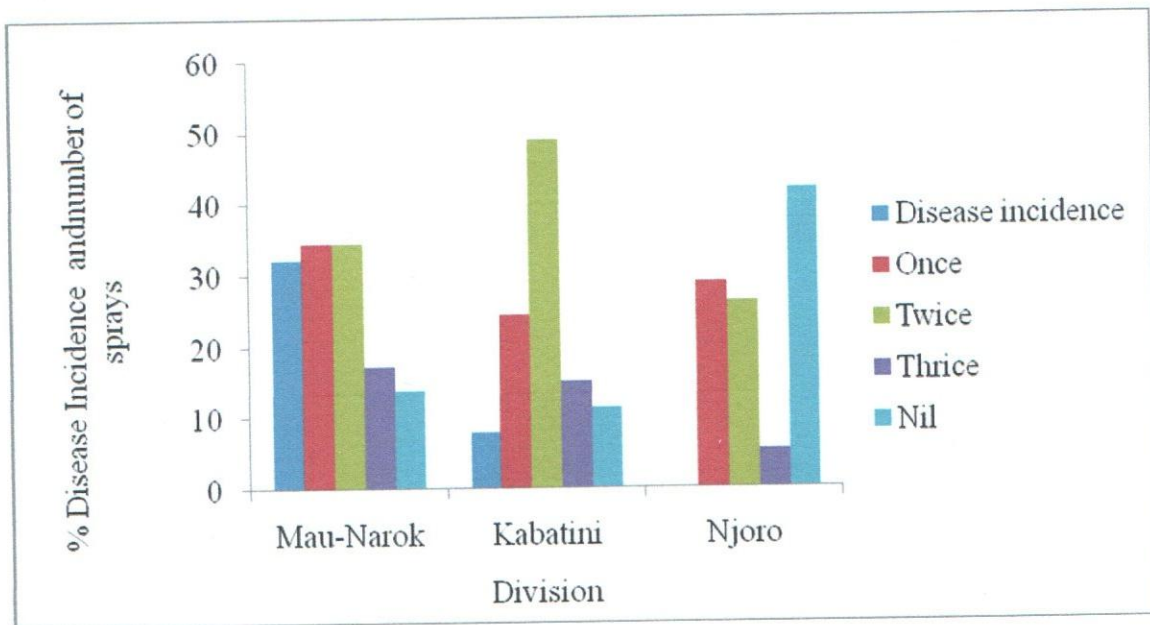


Figure 4.5: Incidence (%) of wheat stem rust disease and number of fungicide sprays per growing season

4.4.4 Effect of Variety and Seed Source on Incidence of Wheat Stem rust

Mwamba variety of certified seed was grown in Mau-Narok division where disease incidence of 23.5% was observed. In the same division the fields with 12.8 % and 13.3% disease incidence were planted with certified seed of the variety Robin. The field with 7.9%, 12.6% and 18.8% disease incidence had certified seed of the variety NjoroBWII. The field with disease incidence at 19.8% was planted with certified seed of Heroe (Figures 4.6 & 4.7). In Kabatini the crop with disease incidence of 3.9%, 11.3% and 9.2% had certified seed of variety Robin. The field with 3.3% disease incidence in the same division was planted with certified seed of variety NjoroBWII (Figures 4.6 & 4.7).

The field that reported the highest disease incidence of 77.8% was found in Mau-Narok division planted with uncertified seed of the variety Mwamba. The same division reported

38.6%, 50.7%, 58.5% and 60.3% disease incidence on the crops having uncertified seed of variety NjoroBWII. The field with 45.8% disease incidence was planted with uncertified seed of variety Robin (Figures 4.6 and 4.7). The percentage of growers using uncertified seed in Mau-Narok was 70.7%, Njoro 23.7% and Kabatini 22.6% (Figures 4.6 and 4.7). The division with many growers using certified seed was Kabatini 77.4%, Njoro 76.3% and Mau-Narok had the least percentage of 29.3%.

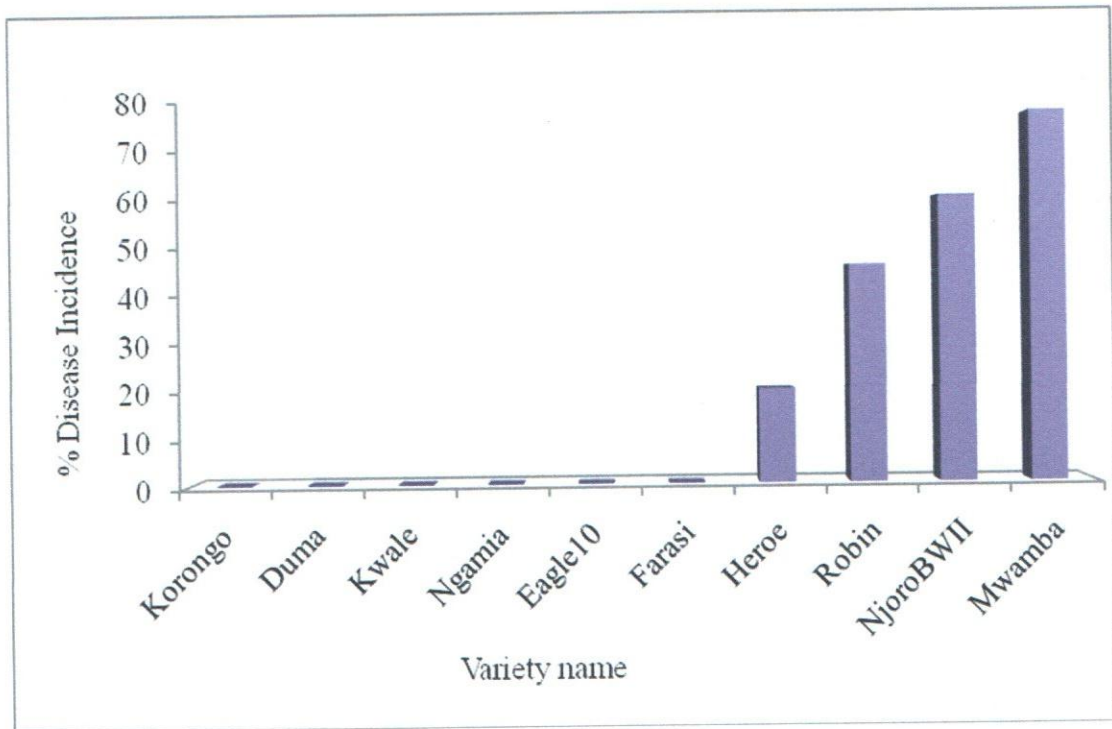


Figure 4.6: Incidence (%) of wheat stem rust disease and variety of uncertified seed

In all the three divisions only bread wheat was cultivated as in the case of most of Kenyan growers. In the Mau-Narok division the growers who planted the variety NjoroBWII was 53.4%, followed by Robin at 27.6%, Eagle10 at 1.7% and Korongo 1.7%. Mwamba was at 5.3%, Kwale 5.2%. The varieties Heroe, Ngami and Farasi were only found in Mau-Narok. In Njoro division the growers with the variety NjoroBWII (23.6%) Robin (34.2%), Mwamba (23.6%), Eagle 10 (2.7%) and Korongo (7.9%). Duma (2.6%) was grown in Njoro division only, Kwale (5.4%). In Kabatini the growers with Robin was (64.2%), NjoroBWII (22.5%), Korongo (5.7%) and Kwale (5.7%) (Figure 4.8).

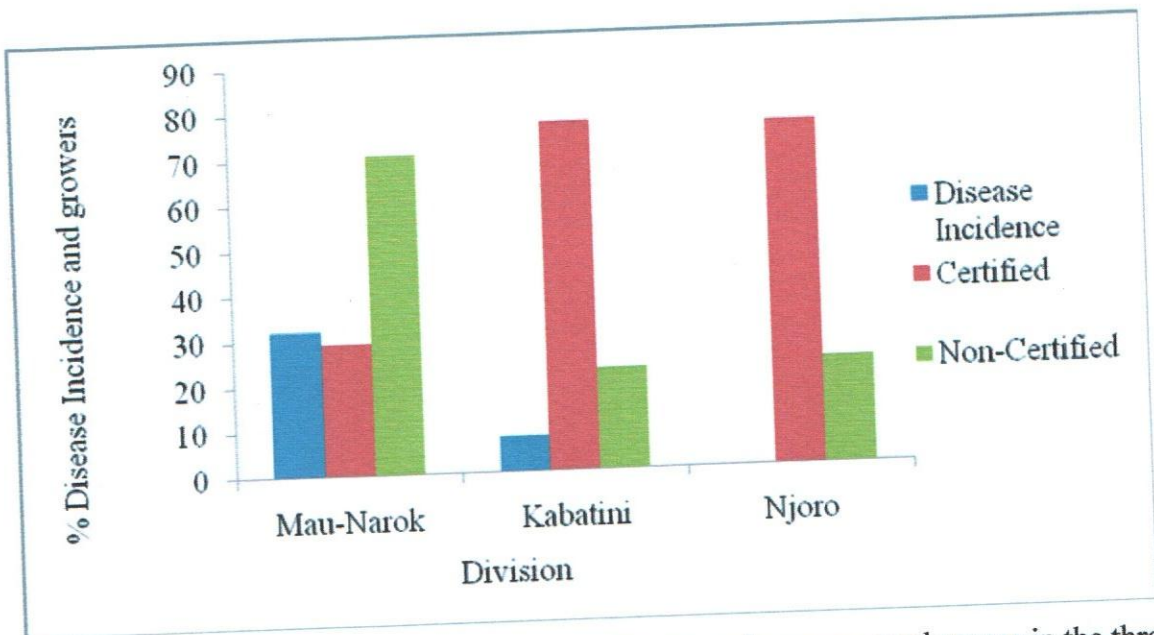


Figure 4.7: Incidence (%) of wheat stem rust disease and growers seed source in the three regions of Nakuru County

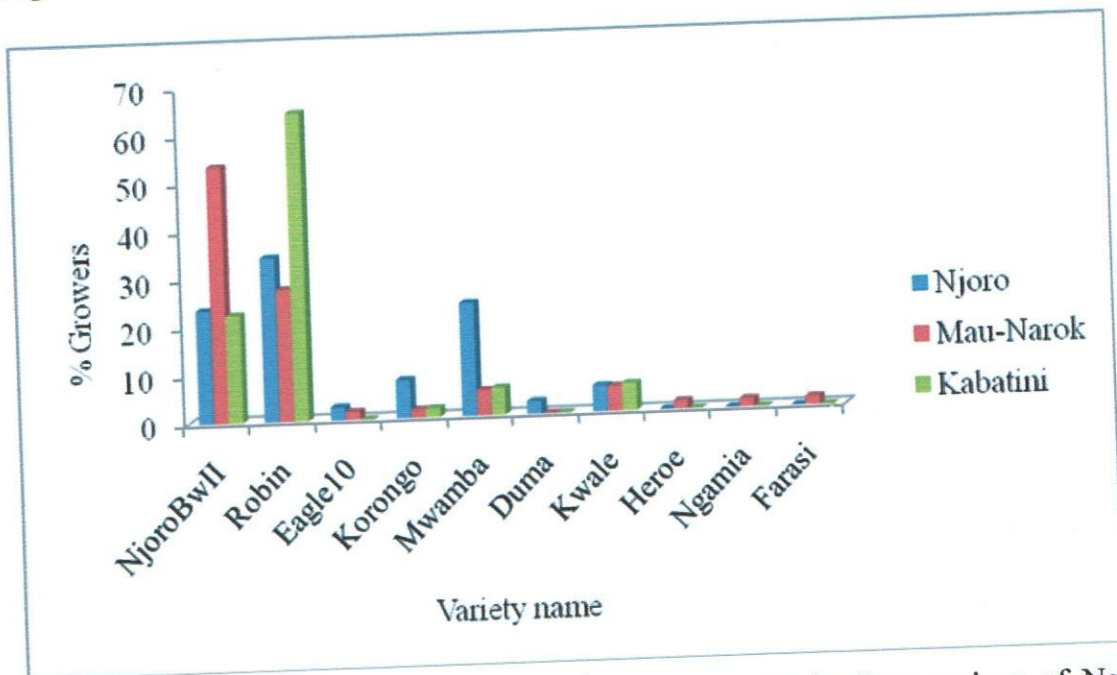


Figure 4.8: Varieties commonly grown and growers (%) in three regions of Nakuru County

4.4.5 Cropping Systems

In Mau-Narok 100% of the growers used a rotation of wheat and peas or wheat and potatoes. In Njoro 94.7% used a rotation of Maize and wheat and 5.3% of growers were found using an intercrop of wheat and Boma Rhodes grass. It was the only division where wheat was

intercropped. In Kabatini 100% used a rotation. The rotation involved wheat, tomatoes or wheat, beans, Kales instead of tomatoes (Figure 4.9).

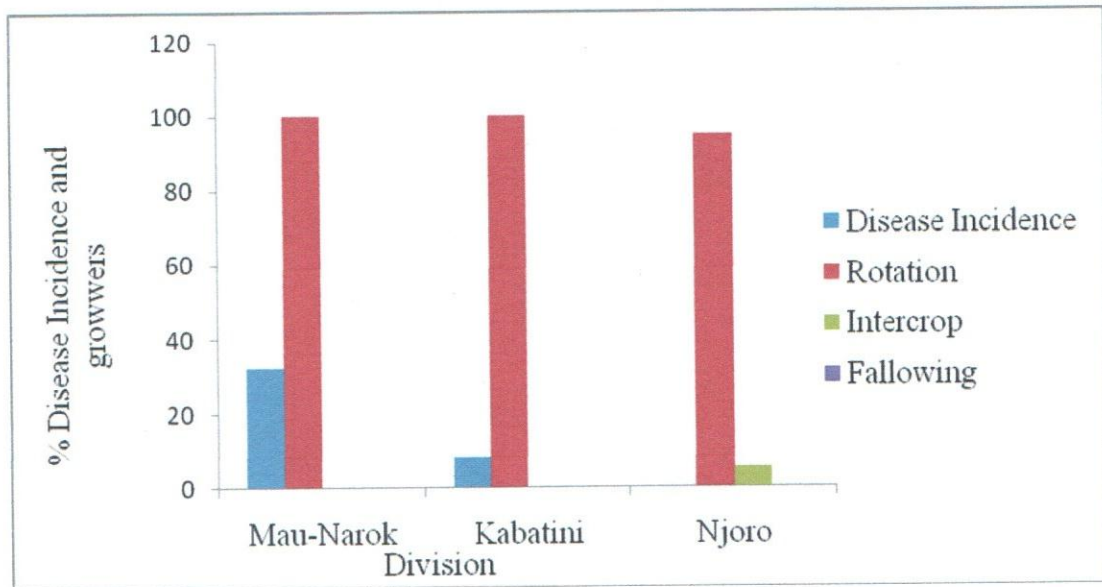


Figure 4.9: Cropping systems and growers (%) found in three regions of Nakuru County

4.5 Discussion

4.5.1 Use of Fungicides in On-Farm Disease Management

The stem rust disease incidence in the three regions showed that there were many factors that were related to disease observed. For the entire study areas incidence ranged from 3.3 to 77.8%. According to FAO-SEC (2012) incidence of over 40% is regarded as high. This implies that in Mau-Narok stem rust disease incidence of 45.6 to 77.8% was high. In Kabatini area the disease incidence was low at 3.3%. The factors that affected disease incidence levels were variety grown, fungicide use such as the rate of spray, number of sprays, seed source and crop management. In the case where of disease incidence of 77.8% in Mau-Narok the crop was weedy, uncertified seed of variety Mwamba was used and one spray was done. The implication is that crop management and production process may affect the level of disease incidence. The fields with disease incidence ranging from 38.6 to 77.8% showed uncertified seeds were used and disease was not controlled using the right recommended spraying regimes. The fungicides used were Nativo and Amistar Xtra made up of a triazole and strobilurin, in the triazole class

were Artea, Folicur, Orius, Silvacur and Topas. Fungicide sprays were done for preventive and curative purposes and none for eradication.

The fields of Kabatini or Mau-Narok with disease incidence ranging from 0 to 12.8% had certified seed of the varieties Mwamba, Korongo, Robin and NjoroBWII were popular. The same fields were sprayed twice or thrice with a fungicide at the recommended rates. However in Mau-Narok where 25.9% of growers sprayed at above the recommended rate was a sign of stem rust disease weighing heavily on growers' management attempts. Considering that the fields were sprayed once or even twice and there was disease incidence of up to 20% is an indicator that management practices have to be made effective.

The stem rust disease severity in all the regions ranged from 20 to 60%. According to Taye, *et al.* (2014), 0-20% indicated low disease severity, 21-40% is medium while greater than 41% is considered as high. Disease severity was high in Mau-Narok, low in Njoro, medium to low in Kabatini. The trend of disease severity was similar to disease incidence. As documented by USDA, (2017) stem rust is favored by hot days 25-30 °C, mild nights 15-20 °C with adequate moisture for night time dews echoed well with diseases escape in Njoro where the temperatures were low and high rainfall. However fungicide use, spraying rates, number of sprays and varieties grown determined the severity levels. The fields sprayed twice or thrice with recommended or above the recommended rate reported medium to low severity of stem rust disease. The number of fungicide sprays also influenced disease incidence or severity Prabhu, *et al.* (2003) reported that two applications of tricyclazole or benomyl controlled panicle blast in rice, as indicated by lower values of disease progress curve and relative panicle blast severity, and increased grain yield. Ganesh, *et al.* (2012) observed that three fungicide applications in rice Tricyclazole or Ediphenphos or Kitazine sprayed thrice at weekly interval managed leaf blast disease in rice. The percent use of fungicides in Mau-Narok was 43.2%, Kabatini 38.9% and Njoro 17.8%. The percentage of fungicide use reflected well with the disease pressure that was being experienced in the three regions with Mau-Narok being the most affected.

Fungicide application as described by Ghazanfar, *et al.* (2009) had an effect on the yield of Paddy rice Rabcide, particularly three applications resulted in increased yield. Gianessi & Reigner (2005) stated that more effective fungicides have been introduced and used by growers to prevent losses caused by fungal pathogens, and Tadesse, *et al.* (2010) also proved that fungicide treatments have effectiveness in reducing disease severity. As stated by Wegulo, *et al.*

(2012) fungicides used to control foliar fungal diseases of wheat belong to two major classes with a broad spectrum of activity against fungal pathogens. Fungicide application by the growers was not clear whether the spraying was done before or after disease onset. As explained by Balardin, *et al.* (2010) fungicide application prior to any contact between pathogen and host is considered to be preventative. After inoculation and just before initial symptoms, the application is curative. All applications made after the onset of symptoms is eradicated. There is therefore a need of fungicide technologies to substantiate on the effective use and control of stem rust disease.

4.5.2 Varietal Use as Disease Management Strategy

All the commonly grown varieties were released as resistant to wheat stem rust but resistance has been breaking down over the years. The two most commonly grown varieties across the three regions were Robin at 41.2% and NjoroBWII at 35.1%. The other varieties Mwamba (10.1%), Duma (5.4%) and Korongo (3.4%) were also found across the regions. High disease incidence and severity were found in the fields with Robin and NjoroBWII which appeared to have become susceptible to stem rust. Generally the fields with Korongo, Duma and Kwale did not report any disease incidence largely due to the number of fungicide sprays used which was twice or thrice as recommended. The low disease incidence could be attributed to genetic resistance which according to Park, (2008) remains the most economical means of rust control. Resistant cultivars also contribute significantly to reducing off-season rust survival. Similarly by Singh, *et al.* (2011) suggested that reducing the area currently occupied by susceptible wheat varieties should become the highest priority.

According to the growers, wheat varieties tend to be replaced for disease management purposes rather than market preference. The two most commonly preferred varieties by the growers in all the areas were Robin (59.1%) and NjoroBWII (40.9%) across the three regions. In contrast varieties Mwamba, Kwale and Korongo were preferred by 10.1%, 5.4% and 3.4% of the farmers respectively. This implied that most farmers preferred old varieties as compared to the newly released varieties. The farmer preference was based on yield and seed quality attributes rather than the disease reaction by the variety.

4.5.3 Cropping Systems and Disease Management

The most common cropping system in the three regions was wheat legume rotation. In Mau-Narok division farmers practiced wheat/peas and wheat potato rotation. The major crop rotation in Kabatini was a rotation of wheat and beans or wheat tomatoes. In Njoro a rotation of wheat and maize was preferred. Overall, 100% of farmers in Mau-Narok practiced crop rotation while 94.7% and 5.3% of the farmers did the same in Kabatini and Njoro respectively. Crop rotation as reported by Khoury & Makkouk, (2010) is one of the most important means of managing disease in small grains. Cultural control methods such as crop rotations, fertilizer use and certified seed not only serve in promoting the healthy growth of the crop, but are also effective in directly reducing disease inocula potential. Besides, crop rotation enhances the biological activities of antagonists in the soil.

Three of wheat fields in Njoro division lower Piave location had an intercrop of wheat and Rhodes grass which according to FAO (www.fao.org) is defined as planting alternating rows of maize and beans, or growing a cover crop in between the cereal rows. FAO (www.fao.org) also reported that the practice is not beneficial because an intercrop may compete with the main crop for light, water and nutrients. This may reduce the grain yields of both crops.

Fallowing was not observed in the three regions. This could be due to the fact that land scarcity is compounded by low soil fertility as was observed by Mwangi (1996). This has resulted in the shortening or elimination of the fallow period without concurrent efforts to increase soil nutrients through fertilizer application or other soil management practices mainly found in Sub Saharan Africa.

4.5.4 Certified and Non-certified Seed Sources

The use of certified seeds of improved varieties is one of the basic factors towards increasing productivity and quality in crop production, consequently raising the income of the farmer an affirmation by Sofijanova, *et al.* (2012). Seed quality is critical for crop establishment and plant vigour. Clean seed ensures field hygiene. About 59.1% of farmers interviewed used certified seed while 40.9% used non-certified seed. Mau-Narok (70.7%) had the highest number of farmers using non-certified seed followed by Njoro (23.7%) and Kabatini at (22.6%) in that order. It was evident that fields with high disease incidence ranging from 45 to 77.8% had non-certified seed. Disease severity ranging from 20% to 60% for found in those farms where non-

certified seed were planted. The fields with certified seed had lower or no cases of stem rust disease as the case in Kabatini where 11.3% disease incidence was reported. Sofijanová, *et al.* (2012) verified that gross margin in wheat production using certified seeds is 36 % higher compared to wheat production using uncertified seeds.

4.6 Conclusion

The differences in stem rust disease intensity using incidence and severity across the sites were determined. The incidence and severity % were high in Mau-Narok, followed by Kabatini which had low levels and Njoro having none. However the disease incidence and severity was associated with the management practices. Two or more sprays of a fungicide at the recommended or above recommended rates showed either no or low disease incidence or severity. In addition the variety grown and seed quality determined disease incidence and severity. Use of uncertified seeds of susceptible varieties increased disease levels.

CHAPTER FIVE

5.1 GENERAL DISCUSSION

Food security in Kenya is still a major concern just like many Sub-Saharan African countries. Diminishing land sizes under wheat and other crops suggest that the situation may be getting out of hand. This signifies that wheat production in Kenya is very essential for attaining food security. Wheat production has to be intensified by growing high yielding resistant varieties with high levels of disease resistance. The two objectives of the study were achieved where genotypes expressing both adult and seedling stage resistance were identified. The disease intensity among the three study areas was assessed and variation was observed.

The breakdown of resistance among commonly grown varieties after a short time is of great concern to the breeders and pathologists. The two most commonly grown varieties NjoroBWII and Robin were reported by the growers as being susceptible to stem rust. The study determined that there are resistant genotypes which may be released or used in breeding programmes. As established by Singh, *et al.* (2011) that breeding resistant wheat varieties that have superior yields than currently grown popular varieties is the best option the same case was reported by Njau, *et al.* (2009) that the most effective approach to prevent losses from stem rust is through the deployment of resistant cultivars. More effective sources of resistance need to be identified and incorporated in the existing commercial cultivars. The clarification by Bingham, *et al.* (2009) that genetic improvement to minimize yield loss under disease is an attractive goal, as it exerts little or no selection pressure on pathogen populations and could form a useful component of durable disease management programme. Therefore constant screening for genotypic resistance for disease, high yielding and stable genotypes is required. There is need for durable resistance because a lot of effort and research funds are required for release of resistant varieties.

Stem rust disease of wheat occurs more in the main wheat growing regions of Kenya, as stated by FAO (2013) these are areas of above 1500 meters in the Nakuru, Uasin Gishu, Trans Nzoia and Laikipia counties. Wheat growers in the regions use inputs for production and mainly fungicide use seemed to be quite an issue. The use of fungicides such as number of sprays per season had growers in some regions such as Mau-Narok spraying thrice and above the recommended rates. The fungicide names such as 'Rustkiller' may imply that growers may be

using the wrong fungicides or taken advantage by dealers. This also means that a lot of effort is required to control and manage the disease in the specified area.

The disease assessment also showed that the disease severity and incidence was high especially in Mau-Narok using parameters such as AUDPC, FDS, and severity and incidence percentages. The study demonstrated that wheat production in Kenya needs to be done with effective management strategies to stem rust, with issues concerning climate change farming practices in place should be done in an ecologically and environmentally friendly manner. The use of an integrated disease management approach may be employed due to the fact that the ultimate goal of the growers is high grain yields. The use of resistant varieties need to be combined with one fungicide spray at most at the recommended rates rather than above recommended. Use of certified seed to maximize on the germination percentage and crop vigour is essential for growers instead of growing other seed either obtained from other growers or farm saved seed.

Fungicide use is useful in control and management of stem rust in wheat as stated by Ivic (2010) fungi develop on the surface of plant organs, providing the pathogen to spread its spores in the environment. The development of fungicides was done as a chemical control for foliar diseases. Fungicide use may be done as a protectant, preventative, curative or eradivative manner but the best method of using fungicides as pointed out by Ivic (2010) that systemic fungicides showed the best effect when applied as preventative measure to diseases infection. There are many fungicide names and brands in the markets and growers should be assisted in the selection and application methods, therefore fungicide technology has to be reviewed and developed constantly.

Use of rotations is meant to improve on the farming practices as affirmed by EU (2012) that it is a cornerstone of a good agronomic practice and sustainable. EU (2012) added that crop rotation has many agronomic, economic and environmental benefits compared to monoculture cropping. Appropriate crop rotation increases organic matter in the soil, improves soil structure, reduces soil degradation, and can result in higher yields and greater farm profitability in the long-term. Increased levels of soil organic matter enhances water and nutrient retention, and decreases synthetic requirements.

5.2 CONCLUSION

There were differences in performance which was highly linked to the pedigrees showing that the ones of the already released varieties were superior. The genotypes with R (Resistance) and MR (Moderately Resistance) were greater in grain yield which translates to resistance is directly proportional to high grain yield. Disease assessment revealed Mau-Narok having high disease pressure which was also revealed when AUDPC was calculated for the three study areas. The farming practices in place such as fungicide use, variety selection, and cropping systems determined the amount of disease occurrence. These suggest that wheat production in Kenya has to be done with effective management options available for stem rust, which may also be applicable in the eastern Africa region.

5.3 RECCOMENDATION

Stability and resistance for both disease resistance and grain yield, across all major growing regions of Kenya is important. Integrated disease management approach for stem rust is the best approach. Verification and validation of an integrated disease management for wheat stem rust is needed.

5.4 FUTURE RESEARCH

The study resulted in the identification of genotypes which are resistant at seedling and adult stage, high yielding and stable across the study areas. More work need to be done on genotypic stability for both disease resistance and grain yield. Identification of genotypic stability across all major growing regions of Kenya requires to be done. The disease intensity across the study areas was revealed which showed disparities across the study areas. The farming practices in place were determined which showed variation also. Work should be done on the verification and validation of an integrated disease management approach for wheat stem rust. Use of resistant varieties, use of certified seed, rotations, general crop management such as weeding. Work needs to be done on verification and validation of an integrated disease management approach for stem rust.

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5.6 APPENDICES

Appendix 1: Description of bread wheat (*Triticum aestivum* L.) genotypes used in the experiment

Genotype	Source	Pedigree/selection history
KSL1	CIMMYT	SER11/CHIBIA/4/BAV92//IRENA/KAUZ/3/HUITES
KSL15	CIMMYT	WBL1*2/BRAMBLING/5/BABAX LR42//BABAX*2/4/ SNI/TRAP31/3KAUZ*2/TRAP//KAUZ
KSL16	CIMMYT	WBL1*2/BRAMBLING/5/BABAX/ LR42// BABAX*2/4 /SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ
KSL17	CIMMYT	BABAX/LR42//BABAX*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP KAUZ/5/WBL1*2/TUKURU
KSL19	CIMMYT	WBL1*2/TUKURU/7/CNDO/ R143/ENTE/MEXI_2/3// AEGILOPSSQUARROSATAUS)/4/WEAVER/5/2* KAUZ/6/FRET2
KSL21	CIMMYT	BW343*2/KUKUNA/3/ SERI//BAV92
KSL22	CIMMYT	PBW343*2/KUKUNA/3/ PGO/SERI//BAV92
KSL13	CIMMYT	QUAIU/5/FRET2*2/4/SNI/TRAP#1/3 /KAUZ*2/TRAP//KAUZ

QUAIU/5/FRET2*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ

CIMMYT

KSL14

Appendix 1: Continued

Genotype	Source	Pedigree/selection history
KSL28	CIMMYT	KFA/5/2*KAUZ//ALTAR84/ AOS/3
KSL29	CIMMYT	TUKURU//BAV92/RA YON*2/3/JUCHI
KSL31	CIMMYT	UP2338*2/KKTS*2//YANAC
KSL32	CIMMYT	UP2338*2/KKTS*2//YANAC
KSL33	CIMMYT	UP2338*2/KKTS*2//YANAC
KSL37	CIMMYT	CAL/NH//H567.71/3/SERI/4/CAL/NH//H567.71/5/2*KAUZ/6/ PASTOR/7/YANAC/8/CAL/ NH//H567.71/3/SERI/4/CAL/NH / /H567.71/5/2*KAUZ/6/PASTOR
KSL40	CIMMYT	CAL/NH//H567.71/3/SERI/4/CAL/H567.71/5/2*KAUZ//PASTOR /7/YANAC/8/CAL/NH//H567.71/3/SERI/4/CAL/NH//H567.71/5/2* KAUZ/PASTOR
KSL46	CIMMYT	TACUPETOF2001/6/CNDO/R143/ENTE/MEXI_2/3/AEGILOPS SQUARROSA (TAUS)/4/WEAVER/5/PASTOR/7/ROLF07
KSL47	CIMMYT	TACUPETO01/6/CNDO/R/R143//ENTE/MEXI2/3/AEGILOPS SQUARROSA(TAUS)/4/WEAVER/5/PASTOR/7/ROLF07
KSL48	CIMMYT	TACUPETO01/6/CNDO/R/R143//ENTE/MEXI2/3/AEGILOPS

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Appendix 1: Continued

Genotype	Source	Pedigree/selection history
KSL50	CIMMYT	WBL1*2/4/BABAX/LR42//BABAX/3/BABAX/LR42//
KSL53	CIMMYT	TILILA/JUCHI/4/SERI.1B// KAUZ/HEVO/3/AMAD
KSL51	CIMMYT	KSW/7/CAL/NH//H567.71/3/SERI/4/CAL/NH//H567.71/5/2 KAUZ/6/PASTOR/8/CAL/NH//H567.71/3 SERI/4/CAL/NH //H567.71/5/2*KAUZ/6/PASTOR
KSL54	CIMMYT	28th SAWSN /09
KSL57	CIMMYT	C 30 SAWSN 2010
KSL59	CIMMYT	C 30 SAWSN 2010
KSL42	CIMMYT	FRANCOLIN #1/KIRITATI
KSL44	CIMMYT	BABAX/LR42//BABAX*2/3/ KUKUNA/4/TAM200/ PASTOR//TOBA97
KSL52	CIMMYT	KENYANYANGUMI/3/2*KAUZ/PASTOR//PBW343
KSL58	CIMMYT	C 30 SAWSN 2010
KSL63	CIMMYT	4th SRRSN 2010

Genotype	Source	Pedigree/selection history
KSL69	CIMMYT	Ethiopia 2010
Appendix 1: Continued		
KSL71	CIMMYT	SOUTHAFRICAN BETHLEHEM2010
KSL72	CIMMYT	4th SRRSN 2010
KSL73	CIMMYT	Bangladesh 2010
KSL76	CIMMYT	K. YOMBI/R1066
KSL81	CIMMYT	NJBW/CHIRIKU
KSL115	CIMMYT	R1071/MBUNI
KSL118	CIMMYT	R1075/KWALE
KSL126	CIMMYT	R1089/R1069
KSL137	CIMMYT	K8676/NJBWII
KSL142	CIMMYT	KONGONI/1083
KSL144	CIMMYT	KWALE/ZABADI
KSL146	CIMMYT	PAKA/R8665
KSL156	CIMMYT	RWAPT60/MBUNI
KSL161	CIMMYT	R960/R1088

Appendix 1: Continued

Genotype	Source
Checks	
Korongo ^a	KALRO
Kingbird ^a	KALRO
Eagle10 ^a	KALRO
Robin ^a	KALRO
Kenya Wren ^a	KALRO

— KSL: Kenyan Selection; CIMMYT, Center for Maize and Wheat Improvement; a: commonly grown varieties

Appendix 2

a) Mean squares derived from analysis of variance for stem rust disease resistance and yield components of wheat genotypes

Source of Variation	d.f	FDS	AUDPC	YIELD	TKW
Rep	2	5068.95	41719.79	6832.39	0.000169
Location	2	7275.17***	688756.54***	2436705.46***	0.001141***
Block (rep)	12	15044.1	241179.9	8313.02	0.0002228
Genotype	49	3046.92***	510471.46***	14311.70***	0.0001528
Genotype*Location	98	27821.88***	33016.39***	12184.86*	0.000098**
Error	10.0	9.17	111.33	64.692	0.0085737
CV%		33.2	36.22	54.57	34.2
R ²		0.89954	0.90194	0.852	0.51773

*, **, ***, Represents significance at $P < 0.05$, $P < 0.01$ and $P < 0.001$ respectively, d. f, degrees of freedom,;

FDS, Final Disease Severity; AUDPC, Area Under Disease Progress Curve, TKW, Thousand Kernel Weight.

CV, Coefficient of Variation, R², Coefficient of Determination

b) Mean square table for AUDPC for the three sites

Source	DF	Squares of squares	Mean Square	F Value	Pr> F
Model	163	32603819.17	200023.43	16.14	<.0001
Error	286	3544805.83	12394.43		
Corrected Total	449	36148625.00			
R-Square	CoeffVar	Root MSE	AUDPC Mean		
0.901938	36.22460	111.3303	307.3333		

c) Analysis of variance table for AUDPC for the three sites

Source	DF	Anova SS	Mean Square	F Value	Pr> F
Rep	2	34688.250	17344.125	1.95	0.1487
Block(rep)	12	1011267.000	84272.250	9.47	<.0001
Genotype	49	6393085.875	130471.140	14.66	<.0001
R-Square	CoeffVar	Root MSE	AUDPC Mean		
0.906693	40.67632	94.34873	231.9500		

d) Mean square table for the Final Disease Severity for the three sites

Source	DF	Sum of Squares	Mean Square	F Value	Pr> F
Model	169	211784.7711	1253.1643	14.89	<.0001
Error	280	23569.7289	84.1776		
Corrected Total	449	235354.5000			
R-Square	CoeffVar	Root MSE	FDS Mean		
0.899854	33.20206	9.174835	27.63333		

e) Analysis of variance table for Final Disease Severity for the three sites

Source	DF	Anova SS	Mean Square	F Value	Pr> F
Loc	2	14550.3333	7275.1667	86.43	<.0001
Genotype	49	149298.9444	3046.9172	36.20	<.0001
Loc*genotype	98	27821.8889	283.8968	3.37	<.0001
Blocks(rep)	14	15044.6515	1074.6180	12.77	<.0001
Rep(Loc)	6	5068.9529	844.8255	10.04	<.0001

f) Mean square table for the grain yield Severity for the three sites

Source	DF	Sum of Squares	Mean Square	F Value	Pr> F
Model	163	24993200.42	153332.52	5.58	<.0001
Error	286	7859089.36	27479.33		
Corrected Total	449	32852289.78			

R-Square	CoeffVar	Root MSE	ACTUALWEIGHT Mean
0.760775	54.09820	165.7689	306.4222

g) Analysis of variance table for grain yield for the three sites

Source	DF	Anova SS	Mean Square	F Value	Pr> F
Rep	2	53110.43	26555.22	0.97	0.3817
Block(rep)	12	812250.88	67687.57	2.46	0.0045
Location	2	16702595.68	8351297.84	303.91	<.0001
Genotype	49	2767804.00	56485.80	2.06	0.0001
Location*genotype	98	4657439.43	47524.89	1.73	0.0003

h) Mean square table for the kernel weight for the three sites

Source	DF	Squares	Mean Square	F Value	Pr> F
Model	163	0.02237668	0.00013728	1.88	<.0001
Error	286	0.02084421	0.00007288		
Corrected Total	449	0.04322089			

R-Square	CoeffVar	Root MSE	KERNEL Mean
0.517728	34.16961	0.008537	0.024984

i) Analysis of variance table for kernel weight for the three sites

Source	DF	Anova SS	Mean Square	F Value	Pr> F
Rep	2	0.00033792	0.00016896	2.32	0.1003
Block(rep)	12	0.00267387	0.00022282	3.06	0.0004
Location	2	0.00228246	0.00114123	15.66	<.0001
Genotype	49	0.00743911	0.00015182	2.08	0.0001
Location*Genotype	98	0.00964332	0.00009840	1.35	0.0300

Appendix 3

i): Area Under Disease Progress Curve (AUDPC) means for the fifty wheat genotypes in the three locations.

Genotype	Lanet	Njoro	Mau-Narok	Means
KSL142	0	25.8	60.8	28.9
KSL71	75	5.8	27.5	36.1
KSL144	0	25.8	82.5	36.1
KSL50	11.7	0	110	40.6
KSL31	60.8	5.8	137.5	68.1
KSL44	33.3	31.7	141.7	68.9
KSL115	33.3	170	72.5	91.9
KSL146	76.7	63.3	165.8	101.9
KSL69	98.3	112.5	140	116.9
KSL76	45	96.7	211.7	117.8
KSL161	88.3	213.3	66.7	122.8
KSL53	94.2	152.5	165	137.2
KSL73	17.5	258.3	151.7	142.5
KSL54	110	217.5	131.7	153.1
KSL51	215	69.2	180.8	155
KSL156	167.5	130	169	155.6
KSL81	141.7	76.7	267.5	161.9
KSL137	66.7	5.8	438.3	170.3
KSL 37	88.3	245	204.2	179.2
KSL72	120	221.7	296.5	212.8
KSL52	157.5	154.2	351.7	221.1
KSL33	82.5	167.5	416.7	222.2
KSL17	120	171.7	385	225.6
KSL57	100	290.8	328.3	239.7
Kingbird	280	295	177.5	250.8
KSL19	150.8	219.2	400	256.7
KSL118	184	219.2	377.5	260.3

Appendix 3:Continued

Genotype	Lanet	Njoro	Mau-Narok	Means
KSL59	106.7	355.8	280	277.5
KSL40	196.7	285	351.7	277.8
KSL22	296.7	335	205.8	279.2
KSL47	251.7	298.3	316.7	288.9
KSL58	160.8	409.2	365.8	311.9
KSL28	204.2	328.3	411.7	314.9
KSL46	185	451.7	316.7	317.8
KSL16	198.3	311.7	486.7	332.2
KSL126	382.5	343.3	345	356.9
Eagle10	480	398	225	367.8
KSL1	210	363.3	544.2	372.5
KSL14	332.5	461.7	396.7	396.9
Korongu	698.3	395	686.7	593.3
K. Wren	530.0	623.3	745	632.8
KSL13	466.7	751.7	721.7	646.7
KSL15	481.7	750	843.3	693.3
KSL29	496.7	805	831.7	711.1
KSL21	710	1231	695	878.9
KSL42	626.7	1080	1040	916.1
Robin	875.8	1093	970	979.7
Average	232	326.7	364.2	307.3

Appendix 3: Continued

ii) Final Disease Severity means for the fifty genotypes in the three locations.

Genotype	Lanet	Mau-Narok	Njoro	Means
KSL142	0	10	0	2.8
KSL71	5	3.3	1.7	3.3
KSL144	0	10	0	3.3
KSL50	3.3	15	0	6.1
KSL31	8.3	16.7	1.7	8.9
KSL44	5	13	1.7	6.1
KSL115	5	11.7	10	8.9
KSL146	8.3	11.7	3.3	7.8
KSL69	11.6	11.7	10	11.1
KSL76	8.3	33.3	8.3	16.7
KSL161	11.7	10	13.3	11.7
KSL53	13.3	21.7	5	16.7
KSL73	5	13.3	20	11.7
KSL54	13.3	15	11.7	13.3
KSL51	16.7	23.3	5	12.8
KSL156	16.7	16.7	11.7	13.3
KSL81	15	30	10	15
KSL137	10	50	1.7	15
KSL 37	11.7	28.3	15	18.3
KSL72	13.3	40	11.7	26.5
KSL52	16.7	43.3	10	18.3
KSL33	10	50	16.7	26.3
KSL17	13.3	46.7	23.3	24.4
KSL57	15	40	11.7	22.2
Kingbird	23.3	13	8.3	15
KSL19	25	36.7	13.3	25
KSL118	30	43.3	15	29.4

Appendix 3: Continued

Genotype	Lanet	Mau-Narok	Njoro	Means
KSL48	20	43.3	13.3	25.6
KSL59	23.3	23	23.3	23.3
KSL40	25.3	46.7	10	26.6
KSL22	36.7	31.7	33.3	33.9
KSL47	30	36.7	18.3	28.3
KSL58	20	53.3	13.3	32.2
KSL28	30	43.3	16.7	30
KSL46	20	36.7	36.7	31.1
KSL16	26.7	46.7	23.3	32.2
KSL126	33	26.7	26	28.3
Eagle10	33.3	16	32.3	27.8
KSL1	28.3	46.7	23.3	32.8
KSL14	40	50	36.7	42.2
KSL63	26.7	50	56.7	44.4
Korongu	53.3	53.3	28.3	45
KSL32	60	63.3	40	54.4
Kenya Wren	50	70	30	57.8
KSL13	53.3	60	60	57.8
KSL15	56.7	66.7	56.7	60
KSL29	46.7	73	66.7	62.2
KSL21	63.3	46.7	90	66.7
KSL42	56.7	73	80	70
Robin	45	80	80	68.3
Average	23.93	23.3	35.7	27.6

i. Papers generated from the thesis

1. Tenge, B. N., Ojwang, P. P. O., Otaye, D., & Njau, P. (2016). Assessment of advanced Kenyan selected wheat lines for resistance to the prevailing stem rust races (*Puccinia graminis* f.sp.tritici) in Kenya. *Journal of Plant Breeding and Crop Science*, 8(7), 94-108.

2. Tenge, B. N., Ojwang, P. P. O., Otaye, D. & Oyoo, E. M. (2016). Wheat stem rust disease incidence and severity associated with farming practices in the Central Rift Valley of Kenya. 11(29):2640-2649.

Appendix 5

i) Questionnaire check list

Part 1 General information

Interviewer: _____

Date of interview: _____ Month _____ Year _____

Questionnaire No _____

1. The division and the location

Regions used

1. Njoro division

Locations

a. Piave

b. Lower Piave

c. Njoro

d. Kerima

2. Mau-Narok division

Locations

a. Sururu

b. Likia

c. Mau-Narok

d. Mwisho wa Lami

3. Kabatini division

Locations

- a. Karunga
- b. Ngecha
- c. Thayu
- d. Ruguru

2. Farm classification

- a. Small scale _____
- b. Medium scale _____
- c. Large scale _____

Part 2 Farming practices

3. Spray of your wheat crop against wheat stem rust Yes No

4. Fungicides did you used

- a. Artea
- b. Silvicur
- c. Folicur
- d. Orius
- e. Nativo
- f. Rust killer
- g. Armester extra
- h. Topaz
- i. Nil

5. Rates of fungicide used

- a. Recommended rates
- b. Above the recommended rates

c. Below the recommended rates

d. Nil

6. Number of sprays

a. Once

b. Twice

c. Thrice

d. Nil

7. The major constraints to pesticide spray

a. Dew

b. Rain

8. The growing seasons in a year

a. Once

b. Twice

9. If wheat grown frequently Yes No

10. Wheat variety grown

a. NjoroBWII

b. Robin

c. Eagle 10

d. Korongo

e. Mwamba

e. Duma

g. Kwale

h. Heroe

i. Ngamia

j. Farasi

11. If variety is replaced Yes No

12 Variety replaced _____

13. How frequently variety is replaced

a. Every year

b. Not replaced

14. Reason for replacement

a. Disease management

b. Market preference

15. The number of years the stated wheat variety is grown

a. one year

b. more than one year

16. Levels of wheat yields obtained

a. High

b. Low

c. Medium

17. If wheat farming is profitable. Yes No

18. used

a. DAP

b. Urea

c. N.P.K

d. S.S.P.

e. Both

f. Booster

g. Nil

19. Cropping system

a. Rotation

b. Fallow

c. Intercrop

d. No rotation

20. Seed source

a. Certified source

b. Non-certified

21. Variety commonly grown

a. Robin

b. Njoro BWII

22. Planting date

a. April

b. May

c. March

d. June

23. Straw disposal

a. Fed to animals

b. Burning

c. Other

24. Where wheat is sold after harvest

a. Brookers

b. Other

25. How wheat is stored after harvest

a. No storage

b. Other

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