EFFECT OF SEED QUALITY AND SOIL BORNE PATHOGEN INOCULUM ON DISEASE PRESSURE IN COMMON BEAN

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DECLARATION

This thesis is my original work and has not been presented for any award of a degree in any other University

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DEDICATION

To my daughter Tiffany N. Wanyama, my husband Evans W. Wafula and in memory of my late father who saw me through my schooling.

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ABBREVIATIONS AND ACRONYMS

AEZ	Agro-ecological zone
ALS	Angular leaf spot
Al	Aluminium
BCMV	Bean Common Mosaic Virus
Ca	Calcium
CBB	Common bacterial blight
CFU	Colony Forming Units
Cu	Coppper
FAO	Food and Agriculture Organization of the United Nations
FAOSTAT	Food and Agriculture Organization of the United Nations- Statistics
GLP2	Grain Legume Program Two
Ha	Hacter
ISTA	International Seed Testing Association
K	Pottasium
KALRO	Kenya Agricultural and Livestock Research Organisation
KK8	Kakamega eight
KK15	Kakamega fifteen
KATX56	Katumani X fifty six
KATX69	Katumani X sixty nine
Kg	Kilogram
LM1	Lower midland zone one
LM4	Lower midland zone four

Mg	Magnesium
ml	Milliltres
Mn	Manganese
MOA	Ministry of Agriculture
MOARD	Ministry of Agriculture and Rural Development
N	Nitrogen
NaCl	Sodium Chloride
NARL	National Agricultural Research Laboratories
NGO	Non- Governmental Organization
P	Phosphorus
Ppm	Parts per million
Zinc	Zinc

ABSTRACT

Build-up of pathogen inoculum in seed and soil due to recycling of seed and continuous cropping by farmers contribute to high bean disease pressure and low yields. This study was carried out to determine the effect of seed quality and soil borne pathogen inoculum on fungal and bacterial disease pressure in common bean. Field experiments were carried out in two agro-ecological zones (AEZs) LM1 (Busia) and LM4 (Bondo). Seeds of four bean varieties, KATX56, KATX69, KK8 and GLP2, sourced from market, farmers and agro-chemical outlets were evaluated for quality before planting and at harvest. During crop growth, data on emergence, stand count, incidence and severity of root rots and foliar diseases was taken at emergence, second, fourth, sixth, and eighth weeks after emergence. Inoculum of soil borne pathogens and soil fertility levels were determined at planting and infections on bean stem bases determined by isolation on agar medium. Plant biomass, number of pods per plant and grain yield was determined at harvest.

Soil nutrient levels varied between AEZ, but all soil samples had below recommended levels in N and C. The soil borne pathogens isolated from soil were *Fusarium solani, F. oxysporum, Rhizoctonia solani, Pythium ultimum* and *Macrophomina phaseolina*. There were variations in the level of root rots inoculum between the two AEZs and sites, with higher populations in soils from LM1 of up to 24,000 CFU/g. Incidence of root rots diseases, infection on stem bases and stand count varied significantly ($p \leq 0.05$) across sites and among seed sources. High disease incidence was in beans grown in LM4 (71%) compared to LM1 (37%). Bean crops planted from certified GLP2 and KK8 had lower root rot disease intensity and infections on stem bases of as low as 15% and 34%, respectively and higher stand count of up to 97%. However, stand count of below 25% was recorded on bean crops raised from farmer saved, market sourced GLP2 and KATX69 with higher disease intensities of up to

90% and infection levels up to 75%. Seed purity, germination and bacterial contamination varied depending on seed source. All seeds except of certified GLP2 and KK8 had purity below the recommended 95% and more than 15% discoloured and shrivelled seeds. Germination of farm saved and market sourced seeds was less than 85%, with high proportions of mouldy seeds and seedling infection of up to 70%. Higher bacterial pathogen inoculum levels of up to 3,187 CFU/seed for *Xanthomonas axonopodis* pv. *phaseoli* and 1,634 CFU/seed for *Pseudomonas savastanoi* pv. *phaseolicola* were detected in farm saved and market sourced seeds compared to certified seeds. Disease intensity and yield varied significantly ($P \leq 0.05$) among crops planted from seed from different sources. Crops raised from farm saved and market sourced GLP2 had higher disease intensity of above 70% and low yields of less than 400kg/Ha compared to30% disease intensity and 1100kg/Ha for certified seeds. The study showed that high disease inoculum levels and poor quality seed are the contributing factors to high disease levels and low yields. Therefore use of certified seeds of tolerant varieties and agricultural practices that improve soil fertility and prevent build-up of inoculum, should be promoted among farmers to achieve optimum bean yields.

Key words: Common beans, farm saved, seed quality, root rots, soil nutrients.

CHAPTER ONE: INTRODUCTION

1.1 Background information

Common bean (*Phaseoli vulgaris)* is the world's most widely cultivated and consumed of all legumes (Acosta-Gallegos *et al.,* 2007; Nga'yu-Wanjau 2013). Common bean is second after maize as a food crop and a major source of protein and food security in East Africa (Sibiko, 2012). In terms of area under production, Kenya ranks first in Africa followed by Uganda and then Tanzania (Katungi *et al.,* 2009; Akibode, 2011; FAO, 2015). Malawi and Ethiopia rank eighth and ninth, respectively according to FAO statistics (FAOSTAT, 2010). However, in terms of production per unit area, Kenya comes second after Uganda (Katungi *et al.,* 2009). Common bean production in Kenya is mainly in highlands and midlands (Ramaekers *et al.,* 2013; Muthii, 2014). The crop is mostly intercropped with sorghum and maize and rarely grown as a sole crop by subsistence farmers (Eden, 2002). Common bean is normally grown twice a year, the first production is during the long rain of between April and June and the second is during the short rain season of between July and October (Wortmann *et al.,* 1998; Atnaf *et al.,* 2013).

Common bean is a popular food to both the urban and rural populations in Kenya. It is also consumed by people from all income levels (Kara *et al*., 2009). Its consumption is high because it is relatively inexpensive compared to meat and play a key role in alleviating malnutrition and other health related complications (Kabutbei, 2014). Consumption of common bean is promoted by health organizations because it reduces the risk of diseases such as cancer, diabetes or coronary heart diseases (Leterme and Munoz, 2002). It is also used as an appetite suppressant to enhance weight loss (Katungi *et al.,* 2009; Kabutbei, 2014).

Production of common bean in Kenya faces a number of challenges which include drought, pests and diseases, shortage of land and seed related problems like lack of high yielding varieties, high price of seed and unavailability of good quality seed (Birachi *et al*., 2011; Namugwanya *et al*., 2014; Kavoi *et al*., 2016). Bean productivity is severely constrained by diseases such as angular leaf spot, anthracnose, root rot and common bacterial blight (Kavoi *et al*., 2016). Scarcity of arable land, aggravated by rapid population growth and unregulated urban expansion (Altieri *et al*., 2012; Sibiko, 2012) has led to continuous cropping and nutrient mining from the soil. Continuous legume cropping and soil infertility as well as soil acidity, are found in the bean-producing areas where use of inorganic fertilizers is very low (Katungi, 2010). All these factors have contributed to buildup of soil borne pathogens in bean growing areas.

Poor access to improved farm inputs for instance certified bean seed and low household income from crop sales has greatly hampered production of common bean (Larochelle *et al*., 2014; Kavoi *et al*., 2016). Farmers rely on informal seed sources whose quality is poor and contaminated with bacterial and fungal pathogens (Almekinders and Kanaal, 2000). Such seed acts as primary source of inoculum and contribute to spread of seed borne diseases (Fininsa and Tefera, 2010). This study was therefore carried out to determine the effect of soil borne pathogen inoculum and seed quality on disease pressure in common bean.

1.2 Problem statement

Despite the economic importance of common bean in Kenya, production has remained low due to a number of biotic, abiotic and socio-economic constraints (Katungi, 2010; Mukankusi *et al*., 2011; Mutale, 2013). Soil fertility, periodic water stress, insect pests and diseases are considered the major agronomic constraints to bean production (Nderitu *et al.*, 1997; Wagara and Kimani,

2007). Angular leaf spot, anthracnose, common bacterial blight, bean rust, bean Common mosaic virus, *Fusarium* wilt and root rots are the most common diseases of beans causing yield losses (Nderitu *et al.*, 1997; Wagara, 2005; Mwangombe *et al.,* 2008). Higher incidence of soil and seedborne fungal and bacterial diseases are associated with common bean seed and soil (Yesuf and Sangchote 2005; Botelho *et al,.* 2013; Mahmoud *et al.,* 2013).

Soil borne diseases due to *Fusarium* spp, *Macrophomina* spp, *Rhizoctonia* spp and *Pythium* spp which act as a complex is a key constraint to bean production in Kenya, causing yield losses of up to 100% (Schwartz, 2011) due to severe rotting of roots especially in favourable environment that limit root growth (Medvecky *et al .,* 2007). Population pressure on land and changes in farming systems such as continuous cropping has led to decline in soil fertility and build-up of inoculum in soil. Higher root rot pathogen inoculum in soil is directly linked to increased incidence and severity of root rots on beans (Bhatti and Craft, 2006).

Lack of good quality seeds due to informal nature of bean seed systems which include farmers' own seed from previous season, bought from the markets or variety exchange among farmers is a major problem in bean production (Buruchara *et al*., 2002; Sperling *et al.,* 2010; Muthii, 2014). This implies that most farmers use uncertified seed that is of poor quality hence pathogen buildup and high disease incidence causing serious challenge to seedling establishment, survival and crop yield (Oshone *et al.,* 2014).

1.3 Justification

Common bean production in Kenya is estimated at 417,000 metric tons per annum (Katungi *et al*., 2011) which is below the annual demand of 500,000 metric tons per annum (Mauyo *et al.,*

2007) prompting bean import from neighbouring countries to meet the deficit (Oshone *et al.*, 2014). Bean yields have been declining over the years for example, yields declined from 600 kg/ha in 1990 to 400 kg/ha in 2004 (MOARD, 2004; Wagara and Kimani, 2007). High population growth rate has put pressure on land hence the decreased unit area under bean production and the need to intensify and increase production per unit area (FAO, 2010).

In sub-Saharan Africa, diseases are considered to be the major threat to common bean production (Akibode 2011; Beebe *et al.,* 2012). The common diseases are Angular leaf spot, common bacterial blight, halo blight, anthracnose and root rots (Koike *et al.,* 2007; Wohleb and Du toit, 2011). Considering the nature of seed borne pathogens in beans, bean seed evaluation is critical as infected seed may have no visible disease symptoms (Yusuf, 2005). Routine seed evaluation and determination of pathogen inoculum levels in soil are necessary diagnostic tools as they allow steps to be taken in management to reduce risk of disease (Nafula, 2008; Joshi *et al.,* 2009). Accurate diagnosis of seed borne and soil borne diseases and their severity is a crucial step towards assessing probable crop losses and taking appropriate management measures. This necessitates the need for proper disease diagnosis and farmers training on management of soil borne pathogens and the importance of using clean seed. Planting disease free seeds on pathogen free soil as components of integrated disease management package in bean production should be promoted among small scale farmers to realize potential bean yields per unit area (Pande *et al.,* 2000; Osdaghi *et al.,* 2009).Therefore, this study aimed at assessing the effect of soil borne inoculum levels and seed quality on disease pressure and productivity of common bean based on on-farm trials in different agro ecological zones and document soil and seed borne disease incidence and severity induced yield gaps.

1.4 Objectives of the study

The overall objective of this study was to contribute to improved bean productivity through use of quality seed and management of bean diseases

The specific objetives were:

i. To determine the effect of soil borne pathogen inoculum on the severity of root rot.
ii. To determine the effect of seed quality on fungal and bacterial disease pressure and yield of common bean.

1.4.3 Hypotheses

i. High soil borne pathogen inoculum increases the severity of root rots disease in common bean.
ii. The quality of common bean seed influences the intensity of fungal and bacterial diseases thus affecting bean yields.

CHAPTER TWO: LITERATURE REVIEW

2.1 Common bean production and distribution in Kenya

Kenya has two production seasons for common bean but majority of farmers grow the crop once a year because of adverse climatic conditions (Katungi *et al.*, 2009). Common bean production in Kenya occurs in Eastern, Central, parts of Rift valley and Western Kenya (MOA, 2013). The crop is produced in a wide range of cropping systems (Oshone *et al.,* 2014). Fifty seven percent of bean production in Kenya is done under multiple cropping systems, mainly in association with maize, banana, roots and tubers, sorghum or millet (Wortmann *et al.,* 1998; Oshone *et al.,* 2014).

Generally, common bean is considered a short-season crop with most varieties maturing in a range of 65 to 110 days from emergence to physiological maturity (Mauyo *et al.,* 2007; Rahman *et al.,* 2014). Maturity period can extend up to 200 days after planting for the climbers. Common bean is a warm-season crop that does not do well in very cold temperatures at any growth stage (Katungi *et al.,* 2009). The crop requires moderate amounts of rainfall (300 - 600 mm) but adequate amounts are essential during and immediately after the flowering stage (Gomez, 2004; Muthii, 2014). The crop is not sensitive to the type of soil but requires a reasonably fertile, well-drained soil and with no conditions that are likely to interfere with germination and emergence (Wortmann *et al.,* 1998). In Africa, crop cultivation is mainly at an altitude above 1000 Meters above sea level (masl), with adequate amounts of precipitation that is above 400 mm and a soil pH that is above 5.5 (Muthii, 2014).

Kenya has an impressive varietal diversity of common bean seed types (Katungi *et al.,* 2009; Balcha and Tigabu, 2015). About 80 different seed types are found in different places of the country but six are most popular. They included: Red and red/purple mottled GLP2 locally known

as Roseccoco, Purple/grey speckled GLP1004 locally known as Mwezi moja, Pinto sugars GLP 92 locally known as Mwitemania, GLP24 also known as Canadian Wonder, GLP585 known as red haricot and GLP 806 known as Zebra (Katungi *et al.,* 2009). GLP2and Canadian Wonder are the most widely grown because of their high yielding attribute though they require heavy rains and high soil fertility for high production (Katungi *et al.,* 2010; Muthii, 2014). Due to problems associated with diseases and poor soil fertility, these varieties are now becoming unpopular (Asfaw *et al*., 2009). This has led to their replacement by varieties like GLP92 and GLP106 that are well adapted to poor soil conditions (Katungi *et al.,* 2009; Mukankusi *et al.,* 2011).

2.10 Farming systems and the agro-ecological zones of Western Kenya

Western Kenya has the most evenly distributed rainfall. The annual averages range between 900 and 2200 mm (Jaetzold *et al.,* 2009). This also stretches towards the lake but rainfall is much lower in Nyanza region because the shore stretches parallel to the trade winds (Jaetzold *et al.,* 2009*)*. The Western corridor of Kenya covers the former Western and Nyanza provinces. The region fall under agro-ecological sub-zones Humid Zone and Lower Midland Zone of between 900 to 1500m above sea level with Lower Midland Sugarcane Zone (LM1), the Marginal Sugarcane Zone (LM2), the Lower Midland Cotton Zone (LM3) and Lower Midland Marginal Cotton Zone (LM4) and Humid Coffee-Tea zone (UM1) (Jaetzold *et al.,* 2009).

Agro ecological zone LM1, the Marginal Sugarcane Zone, has a growing period of 215 days. The annual average temperature is between 21.0 and 22.7 °C. Humidity is relatively high due high temperature and the expansive lake. The annual potential evapo- transpiration is 1 800 - 2 030 mm. The AEZ has two rainy seasons, long rainy season and short rainy season. The first rainy season

occurring between March and May is well suited for crops like medium maturing maize H622, 623, late maturing sorghum like E1291, Finger millet, rice, medium maturing beans like caurentino and pigeon peas. The second rainy season , starts towards the end of August is suited for crop like low land early maturing five series maize, medium maturing sorghum, cowpea, soya bean, sunflower and Robusta coffee (Jaetzold *et al.,* 2009).

Agro ecological zone LM4, the Marginal Cotton Zone is characterised by medium to short cropping season and an uncertain weak second rainy season. It receives rainfall of 400 to 900mm during the first rainy season and less than 150 to 800 mm during the second rainy season and a growing period of 130 days, only 45 days are fairly sure. The first rainy season begins in midMarch. Early maturing sorghum like Serena, early maturing foxtail or proso-millet beans like Katheka, green grams, cowpeas, chick pea, early maturing sunflower and whole year sisal are the crops suitable for this zone (Jaetzold *et al.*, 2009).

2.2 Importance of common bean

Common bean (*Phaseolus vulgaris*) is an important crop for staple food and income generation in Kenya (FAO 1999; Ochilo, 2013) whereby about 417,000 metric tons are produced yearly (FAO STAT, 2010). The contribution of common bean to food security and nutrition in Kenya is relatively high compared to other pulses (Katungi *et al.,*2009; Akibode, 2011*)*). Per capita consumption is estimated at 14 kg per year, which can be up to 66 kg per year in parts of Western Kenya (Mauyo *et al.,* 2007; Katungi*,* 2010). Common bean ranks second to maize as a staple food crop to more than three million farm households in Kenya (FAO STAT 2010; Katungi *et al*., 2015). Beans are an important source of protein, calcium, energy, folic acid, dietary fibre and carbohydrates (Mauyo *et al.,* 2007). They also contain lysine, a nutrient that is relatively deficient

in most staple diets and a good complement to maize, rice, vegetables, banana, cassava and potatoes (Muthii, 2014). Beans have low cholesterol and fat content therefore its consumption reduces cancer, diabetes and heart diseases (Kabutbei, 2014).

Common bean is important for providing multiple forms of food supply as leaves, pods, green grains and dry beans. It is consumed as boiled green leaves, green immature pods and/or dry grains (Katungi *et al.,* 2009). However, fresh beans are difficult to keep, and as such, majorly consumed as cooked or boiled as dry grain. It can be prepared in a wide range of recipes, mixed and cooked with maize locally known as *Githeri* or cooked and mashed alone to form a paste and served with other stew (Katungi *et al.,* 2009).

Common bean generates cash income to households used to purchase other foods and other household needs when other crops have not yet matured (Legesse *et al.,* 2006; Sibiki, 2012). The income generating aspect of bean production is becoming more significant principally near urban market where population relies on beans as an affordable source of protein (Namugwanya *et al.*, 2014). A recent economic survey by the East Africa Bean Research Network's (EABRN) indicated that nearly 50% of bean producers sell part of their harvest, mainly to urban areas where consumption is high (Katungi*,* 2010). The canning type is primarily grown for export market in other neighbouring countries particularly Ethiopia (Oshone *et al.,* 2014).

Common bean is a low input crop that is very useful when incorporated at flowering stage as green manure to help improve soil nutrition (Muthii, 2014). Bean, like other legumes, help in fixing atmospheric nitrogen (Amannuel *et al*., 2000). Dry threshed residues and stalks can be used as mulching materials and also used as livestock feeds (Muthii, 2014). In other cropping systems,

beans serve as component of crop rotation, with cereals, brassicas or solanaceae crop families reducing soil pathogens (Muthii, 2014; FAO, 2015).

2.3 Constraints in production of common bean

For the last few years, production per unit area of common bean in Kenya has been decreasing (Katungi *et al.,* 2010) due to biotic and abiotic constraints (Wagara and Kimani, 2007). Drought, characterized by inadequate, erratic rainfall distribution and long dry spells has significantly reduced yield (Katungi *et al.,* 2009). Inadequate rainfall accounts for over 50% of the total bean yield loss in Kenya (Katungi *et al.,* 2009; Birachi *et al*., 2011).

Declining soil nutrients due to continued nutrient mining without replenishing is also a major constraint that has led to low bean yields (Kajumula and Muhamba, 2012). Deficiencies of nitrogen (N), phosphorous (P), potassium (K), magnesium (Mg), zinc (Zn), and calcium (Ca) and manganese (Mn), aluminium (Al) and salt (NaCl) toxicities (Allen *et al.,* 1996; Wortmann *et al.,* 1998) have all affected beans production. Kajumula and Muhamba (2012) working on bean adaptation to low phosphorus, estimated that beans remove 12.5kgP/ha which is higher than the phosphorus fertilizer additional to soil by resource-poor farmers. Low use of fertilizer due to high costs and lack of enough organic manure are the challenges faced in improving soil fertility (Katungi *et al.,* 2010; Kajumula and Muhamba, 2012; Muthii, 2014).

Low seed availability, inaccessibility and lack of information about market, regional suitability and adaptability, have made adoption of new high yielding varieties slow (Kavoi *et al*., 2016). Thus most farmers opt to obtain seeds from other sources which are of poor quality (Katungi *et al.,* 2009). Small scale farmers routinely plant farm saved bean or seed bought from the market

that is of poor quality and has low resistance to biotic stresses. This is due to high cost of certified seed and lack of awareness on availability of improved bean seeds (Kavoi *et al.,* 2010).

Pests and diseases are the most destructive and most damaging to seeds, leaves, pods and roots (Gomez, 2004). The insect-pests include foliage and pod pests such as the bean stem maggot, striped bean weevil, foliage beetle, black bean aphid, common whitefly, leafhoppers, flower thrips, red spider mites, pod and seed feeders, legume pod borer, cotton bollworm (*Helicoverpa armigera),* and bruchids (Birachi *et al*., 2011; Wanjau, 2013). Losses due to the disease are as a result of premature defoliation, shrivelled pods and shrunken seeds. (Katungi, 2010; Nga'yu-Wanjau, 2013) Common bean is also affected by parasitic nematodes such as the root knot nematodes (Wanjau, 2013). These diseases have made some susceptible varieties to be abandoned or neglected. Management and control has also been costly and difficult to poor farmers.

2.4 Diseases of common bean

Bean diseases are considered one of the major agronomic constraints in bean production in Africa (Allen *et al.,* 1998; Katungi, 2010). Diseases such as angular leaf spot, anthracnose, rust, common bacterial blight, bean common mosaic virus (BCMV), *Fusarium* wilt and root rots are devastating to bean production (Nga'yu-Wanjau, 2013).

Bean anthracnose caused by *Cholletotrichum lindemuthiunum* is one of the most destructive diseases of common bean worldwide. The pathogen causing the disease is seed borne (Mohammed, 2013) and has high pathogenic variability with more than 100 different races (Giraldez *et al.,* 2011). The disease causes 70-76% losses under favourable conditions that are high moisture and low temperatures. The disease causes darkening of the veins on the underside of leaves and angular veins on the upper surfaces of the leaf. Lesions on stems and pods are crater-like and dark brown.

In wet weather, light orange masses of spores are produced in the centre of the lesions leading to secondary spread by rain splash (Mohammed, 2013; Amin *et al.,* 2014). Infected seed may exhibit dark brown spots or rings of discoloration (Allen *et al.,* 1996). On seedlings, the fungus produces necrotic lesion on cotyledons, hypocotyls and on petioles.

Angular leaf spot disease caused by *Phaeoisariopsis griseola,* is one of the most destructive and widely distributed diseases of common bean causing yield losses as high as 80% (Schwartz *et al.,* 1981; Nga'yu-Wanjau, 2013; Pamela *et al.*, 2014). The primary source of infections is the bean seed but other sources include; plant debri, volunteer crops and off season bean crop (Pamela *et al*., 2014). The disease causes premature defoliation, shrivelled pods, and shrunken seeds. A survey conducted in Kenya on angular leaf spot showed a prevalence of between 65-80% (Mwang'ombe *et al.,* 2007). According to Pamela *et al*. (2014), every 10% increase in ALS severity results in 7.9% yield loss. The disease was found prevalent at an altitude ranging from 963 to 2300 m above sea level. Hence ALS is severe and highly prevalent in Kenya, spanning across all the agroecological zones and altitudes where common bean is grown.

Bean root rots is caused by a complex of fungal pathogens mainly *Fusarium* spp *Macrophomina* spp, *Pythium* spp and *Rhizoctonia* spp. *Fusarium* spp is found in the soil and can survive for many years causing infections on the bean crop. The pathogen is favoured by soil acidity in poorly fertilized soil and hot weather and can survive in soil for years (Naseri, 2014). *Macrophomina phaseolina* has a wide host range affecting more than 500 plant species in the world (Reetha *et al.,* 2014). *Macrophomina* is commonly known to cause root and stem rots variously referred to as charcoal rot, dry root rot and ashy stem blight (Fuhlbohma *et al.,* 2013). The pathogen produces macrosclerotia and pycnidia which oozes out conidia. The conidia and macrosclerotia produced

can be disseminated in seed, soil and plant residue as primary inoculum (Songa and Hillocks, 1998; Fuhlbohma *et al*., 2013).

Pythium ultimum is responsible for causing damping off and root rots disease in legumes. The pathogen produces spherical sporangia containing structure and oospores which act as the survival structure and primary inoculum (Lodhi and Khanzada, 2013). *Rhizoctonia solani* infects the root tissues by means of sclerotia or mycelia which survive in the soil for longer periods to cause damping off (Strausbaugh *et al*., 2011). Favourable weather conditions such as high humidity, lead to formation sclerotia which are global shaped (Strausbaugh *et al.,* 2011). Symptoms of root rots include stunting, yellowing of leaves, brown discoloration of the tap root system, damping off and wilting (Schwartz, 2011).

Bacterial diseases of common bean are widespread and distributed throughout bean growing areas with favourable conditions that are moist and warm (Akhavan *et al*., 2013). Bacterial diseases include halo blight caused by *Pseudomonas savastonoi* pv. *phaseolicola* and common bacterial blight caused by *Xanthomonas axonopodis* pv. *phaseoli*. Halo blight causes high yield losses under favourable conditions of cool temperatures, high moisture and plant wounding especially caused before flowering (Osdaghi *et al*., 2009). The pathogen survives in bean seeds and residue for more than four years (Arnold *et al.*, 2011). The disease is characterised by small water soaked lesions on leaves. This later turn to greenish yellow halo and it results to death of new foliage. Pods, stems and petioles also exhibit water-soaked lesions and sometimes producing white or silver colored bacterial ooze (Allen *et al.,* 1996). Pod infection causes seed discoloration and shrivelling.
Systemic infections can lead to severe chlorosis. Bacterial contamination of seeds reproduces seedlings that appear rotten at the nodes (Allen *et al.,* 1996).

Common bacterial blight is seed borne bacterial disease. The primary source of inoculum for CBB, like other bacterial seed-borne diseases is the infected seed and debris (Mehrotra *et al.*, 2003; Akhavan *et al.,* 2013). The initial symptoms are water soaked spots, angular leaf lesions that make the tissues appear scalded (Osdaghi *et al*., 2009; Akhavan *et al.,* 2013). The lesions later expand and merge into dark brown irregularly shaped lesions surrounded by a narrow yellow halo (Allen *et al,.* 1996). The water soaked round spots on pods exudes bacterial ooze (Koike *et al*., 2007). These spots eventually dry and appear as reddish brown lesions. The disease is favoured by worm wet conditions and overhead irrigation which facilitate spread to cause severe infection (Akhavan *et al.,* 2013)

Ascochyta blight is very severe disease of cool seasons and can survive and reproduce in and spread from crop debris or be transported in infected seed. Ascospores are disseminated by wind from the debris as primary inoculum and secondary cycles are initiated by conidia spread by rain splash from plant lesions. The fungus is host-specific in causing disease but may be able to survive in non-host plants and reproduce on their debris. Seed certification is the primary means of preventing its spread to new areas and the importation of new genotypes of the fungus to areas already infested.

Viral diseases are also some of the constraints to bean production in Kenya. One important such disease is common mosaic virus disease caused by bean common mosaic virus (Cannel *et al*., 1979; Mavrič *et al*., 2004) and bean yellow mosaic virus. The virus is seed borne and can be transmitted by several aphid species (Mukeshimana *et al*., 2003). It is of economic importance as it leads to severe crop loss (Sengooba *et al.,* 1997).

2.5 Management of common bean diseases

Bean diseases are usually introduced to a production field mainly by infected seeds and farm machinery during cultivation or harvesting (Souza *et al.,* 2012). Management strategies used to minimize seed-borne and soil borne disease infection in the field include host resistance, cultural, chemical and biological control methods

Cultural management is achieved by removal and burying of infested bean debris in the soil after harvesting to reduce overwintering inocula. Seed storage facilities and agricultural materials should be disinfected (Mohammed, 2013). Production of disease free seeds in semi-arid areas, where conditions are not favorable for disease infection is vital (Yesuf and Sangchote, 2005). A two-year crop rotation with non-host is highly recommended to minimize initial inoculum (Gan *et al.,* 2006). Scouting the fields weekly for symptoms of disease is recommended so that seeds from infected plants are not harvested (Batureine, 2009). Ensuring adequate plant spacing and weed control promotes proper air circulation and decrease moisture in the foliar canopy (Bush, 2009). Physical methods like soil solarization through covering the soil with transparent plastic sheeting for one month before sowing results in reduction of diseases inoculum levels in soil (Mohammed, 2013). A hot-water seed treatment by soaking at 32 to 40°C for 15 hours followed by another soaking at 85° C for 25 minutes has been reported to kill most fungal pathogens in infested seeds without reducing germination (Bush, 2009).

Biological control has proven to be strongly effective against most seed borne infection. Studies have shown that smearing seed with spore suspension of *Trichoderma viridae* reduces infections by *Colletotrichum lindemuthianum (*Mohammed, 2013). Soil drenching with *Trichoderma harzianum* also greatly reduces soil borne inoculum levels. A strong local protection against bean

anthracnose and *Fusarium* spp has also been obtained when susceptible bean leaves are treated with *Trichoderma harzianum* in a liquid medium (Mohammed, 2013). Smearing infected seeds with *T. harzianum*, *T. viridae*, *T. hamatum* and *Gliocladium virens* for 15min and drying them overnight before sowing significantly reduces infection of most fungal diseases and increases seed germination (Sharma *et al.,* 2008). The main antagonistic activities of these bio-agents are through, mycelial growth inhibition, toxic volatile metabolite production and inhibition of spore germination (Anitha *et al.,* 2001). Some antagonistic micro-organisms e.g *Bacillus* and *Streptomyces* species produce antibiotics that have the potential of controlling diseases like bean rust when incorporated in integrated disease management programs (Wagacha *et al*., 2007).

Botanical plant extracts such as neem, garlic, labill, turmeric and ginger have effectively been used to control bean diseases (Mohammed, 2013). Studies by Chandel and Sharma (2014) on efficacy of plant extracts on *Rhizoctonia* spp reported that neemgold or neemazal and cruciferous residues particularly of cauliflower and cabbage (2g/kg of soil) can effectively be used in managing stem rot caused by *Rhizoctonia* spp. Neem (*Azadirachta indica*) seed extract effectively inhibited both germination of conidia and mycelial growth of most fungal pathogens like *C. lindemuthianum* (Mohammed, 2013). Oils such as nettle oil, eucalyptus oil, lemon grass oil and thyme oil have been effective in reducing infection by viruses (Gurjar *et al.,* 2012). Secondary metabolites such as phenols quinones, flavones, flavonoids, flavonols have shown antimicrobial effect and serve as plant defense mechanisms against pathogenic microorganisms (Chandel and Sharma, 2014). A study by Mokhtar *et al*. (2014) showed more than 70% reduction in root rot disease incidence when compost and Eucalyptus or cabbage residue combined is used.

Host plant resistance is the most effective and efficient method of disease management (Pamela *et al.,* 2014; Ssekandi *et al.,* 2015). However, this has been complicated by the presence of several forms or races of most pathogens, and the fact that plants resistant to one race may be susceptible to another. Although planting resistant cultivars is the most effective, least expensive, and easiest for farmers to adopt the possible breakdown of resistance due to adaptation of the pathogen is the main drawback for its application (Schwartz and Singh, 2013)

Chemical management using fungicide in common bean production is extremely rare. However, various fungicides are routinely used to control some diseases like and angular leaf spot and rust (Liebenberg and Pretorius, 2010). For example, use of the triazole and tebuconazole triples yields under conditions of high disease pressure (Pamela *et al.,* 2014). Use of chemical for seed dressing at recommended rates have proven to be the most effective in controlling bean diseases (Mohammed, 2013). High pathogenic variability and new races of pathogens are reported frequently. Thus, integrated disease management is considered to be the most effective approach to minimize yield losses due to diseases. The integration of soil solarization, seed treatment and foliar sprays at the recommended rates are effective in reducing bean disease epidemics (Mohammed, 2013; Kadaari, 2015). Botanicals and bio pesticides formulations along with fungicides have been evaluated and gave promising results.

2.6 Importance of soil borne pathogens in bean production

Root rots are widespread and cause significant losses up to 100% especially in poor soils (Gichuru and Gathoni, 2008). Root rot pathogens include *Fusarium solani* f sp. *phaseoli*, *Pythium ultimum*, *Rhizoctonia solani* and *Macrophomina phaseolina.* The inoculum of *Fusarium* spp can survive for many years in soil . The pathogen is favoured by hot weather, soil acidity and poorly fertilized

soil to cause rotting of roots in bean plant (Naseri, 2014). *Pythium ultimum* and *Rhizoctonia solani* cause damping off to a susceptible host, under humid conditions, warm temperature and cool wet soil condition (Strausbaugh *et al.,* 2011; Lodhi and Khanzada, 2013). *Macrophomina phaseolina* has a wide host range and thrives well under high moisture level in soil and heat stress (Gautam *et al.,* 2014). General symptoms include poor seedling establishment, damping-off, stunting and uneven growth, chlorosis, premature defoliation, death of severely infected plants, poor stand and lower yield (Abawi and Ludwig, 2010).

Roots of infected plants are reduced in size, discolored, and exhibit various stages of decay. The tap root of severely infected plants often die, but large numbers of adventitious roots are produced from the stem above the infected areas and near the soil surface (Otsula *et al.,* 2003; Nzungize and Lyumugabe, 2012). Specific damage symptoms and diagnostic lesions of distinct shapes and colors are produced on infected roots and/or stems when beans are attacked by a single pathogen (Abawi and Ludwig, 2010; Baudoin *et al*., 2013). The involvement of multiple pathogens with diverse biology in causing root rots of beans has made it difficult to effectively control these diseases with a single and practical management option (Kadaari, 2015).

2.7 Management of soil borne pathogens

Management of soil borne pathogens has been difficult due to the presence of survival structures such as sclerotia, mycelia and oospores in soil for many years acting as a source of inoculum for subsequent seasons (Rani and Sudini, 2013). Some of the disease management strategies practiced by farmers include cultural, biological, chemical control and host resistance (Mazzola and Reynolds, 2010; Nzungize and Lyumugabe, 2012). Integrated management strategies have been developed resulting in reduction of inoculum levels. Cultural practices which include fertilizer

application, crop rotation, tillage practices, intercropping and addition of soil amendments improve soil fertility which indirectly reduces the population of soil borne pathogens and disease severity (Abawi and Wildmer, 2000; Martin, 2003). Biological control using microbes such as *Bacillus subtilis Trichoderma* spp and *Pseudomonas flourescens* balances the pathogenic populations through antagonism (Mokhtar and El-Mougy, 2014). Chemical control in form of soil fumigants, seed treatments drenching and foliar sprays with fungicides greatly reduces oomycetes fungal pathogens (Rani and Sudini, 2013).

2.8 Importance of seed quality in bean production

Quality bean seed is an important component to improved bean productivity (Rubyogo *et al.,* 2005). Poor quality seed limits the potential yield of beans through spread and increased severity of diseases (David and Sperling, 1999). There has been the need to constantly improve seeds through breeding and selection of disease resistant common bean germplasm (Katungi, 2010). Drought tolerance and tolerance to low soil fertility is also emphasized in Kenya because of declining soil fertility and drought conditions. During seed production, quality seed is enhanced through seed certification program that involves field inspection of seed crop (Rubyogo *et al.,* 2005; Muthii, 2014). International Seed Testing Association (ISTA) has set standards for certification of bean seed at 99% varietal purity, 0.95% maximum inert matter, 0.05% maximum other seeds, 85% minimum germination and 14% maximum moisture content (ISTA, 1999). These seed requirements guide seed assessment on quality status of farmer seed sources (David and Sperling., 1999; Muthii, 2014).

Damages due to seed-borne pathogens include seed abortion, reduced seed size, shrunken seeds, seed rot, seed necrosis and seed discolouration, all considerably affect seed quality (Muthii, 2014).

Planting farmer saved seed that is untreated and poor quality seed encourages the spread of seedborne pathogens resulting in build-up of inoculum which could eventually lead to outbreak of disease epidemics (David and Sperling, 1999; Opole *et al.,* 2003).

2.9 Bean seed systems in Kenya

The available bean seed systems in Kenya are the formal, informal and semi-formal seed systems. The formal seed system in Kenya is regulated by the governments and private or industry players. It provides new and improved varieties of certified seeds of relatively good quality and high purity. This sector produces only 2% of the bean seed requirements as it only accommodate fewer already popular and widespread varieties (Rubyogo *et al*., 2007). Development of indigenous varieties is not given sufficient attention through the formal system (CTA, 2014). seed under this system are very expensive costing 2 to 4 times the cost of obtaining the seed from the local markets and yield gains not measuring up to the costs of acquiring the certified seed (Zerbe, 2001). High disease management associated with centralised seed production with farmers expected to meet all these costs make the seed more expensive (Sperling *et al.,* 1992). In addition very minimal promotion on consumption of formal seeds is done by the stockist or the agents therefore farmers lack the information on their availability as well as the benefits of using such seed (Rubyogo *et al.,* 2007).

The informal or traditional seed system is semi-structured and operates mainly at the individual or community level. Informal channels which include farm saved seeds, seed exchanges with neighbours and local grain or seed market is more preferred (Oshone *et al.,* 2014). It contributes up to 80% of the total bean seeds used in Kenya (Rubyogo *et al.,* 2007). This is because it makes the seed more accessible and readily available and is more affordable to most farmers than formal seed. This system supplies several genotypes well adapted to the local ecology and farming

practices. Farmer saved seeds predominate the seed sector. This system lacks adequate support, knowledge, skills and incentives for self-regulation and attracting private sector investment, the inadequate linkages to sources of improved bean varieties, inadequate quantity and quality of seeds are among the challenges of this system (Sperling *et al.,* 1992).

The semi-formal seed system is a blend of the formal and informal systems. The systems focus on improving local varieties through breeding and seed selection and introducing improved seeds from national agricultural centres, and International Agricultural Centres. The systems make use of the large variety of both locally improved crops as well as seeds of improved varieties released by the formal systems. It operates between the formal and informal seed systems as they can introduce both improved varieties as well as proven, quality declared seeds of local varieties. Quality declared seeds are sometimes referred to as standard seeds where regular inspections are conducted for diseases and pests. These systems can also produce certified seeds through the use of formalized and standardized quality control measures using small scale seed enterprises, and integrating them into seed markets. The systems can also improve on the informal systems to produce standard seed and/or quality declared seeds, for exchange within the community (Endeshaw *et al*., 2010).

Farmers and many other actors such as NGOs, research institutions, seed parastatals have taken interest in the informal seed system mainly because it is a low cost source of seed, reliable, efficient and accessible channel to provide resource-poor farmers with seeds of improved varieties (Endeshaw *et al*., 2010). Such an interest in the informal seed sector was triggered by the limitation of the formal seed sector to deliver affordable seeds of different crop varieties to the diverse farming community (Endeshaw *et al.,* 2010). Studies have indicated that farmers prefer their own

saved bean seed due to lack of money to buy seed and seed scarcity during planting season (Oshone *et al.,* 2014). Using their own seed also enables them to maintain preferred varieties and good quality seed (Scott *et al.,* 2003).

Local markets are an important source of bean seed for farmers, mostly for the poor who depend on bean for food and income (Scott *et al*., 2003; Rubyogo *et al*., 2005). They consume or sell most if not all the seed during periods of acute food shortages or favourable market prices and therefore the need to supplement farm saved seed (David and Sperling, 1999). Few farmers are normally seed sufficient at the beginning of planting season while majority of farmers rely on off-farm seed. They do so to replenish their stock after crop loss, an emergency, after consuming or selling off existing stock, to expand bean area under production or to obtain new varieties (Katungi *et al*., 2009). Poverty related factors have also contributed to the gap between bean production and high per capita consumption and utilization. All these have forced resource-poor farmers to rely on poor quality off farm seed sources during planting season (Katungi *et al.,* 2009).

Neighbour's seed is locally-adapted varietal mix and generally well-sorted, seed is costly and limited in availability (Sperling *et al.,* 2010). Many farmers therefore go to the local markets to obtain their seed (Scott *et al.,* 2003).

CHAPTER THREE
EFFECTS OF SOILBORNE PATHOGEN INOCULUM ON SEVERITY OF ROOT ROT IN COMMON BEAN

3.1 Abstract

Production of common bean in Kenya has been declining due to root rot caused by a complex of soil borne pathogens. The disease causes up to a 100% yield losses under favourable conditions. Field experiments were carried out during 2015 long rains in Busia (AEZ LM1) and Bondo (AEZ LM4) to evaluate the effect of soil borne pathogen inoculum levels on severity of root rot. Soil samples were collected at planting and analysed for nutrients and inoculum levels. Treatments were four bean seed varieties: KATX56 from markets, farm saved KATX69, Certified KK8 from KALRO, GLP2 from market, certified GLP2 from agro stockist and farm saved GLP2. Other data collected included stand count at emergence, root rots incidence, and seedling infections with root rot pathogens. Soil samples had low levels of total N and C while Samples from Butula had low levels of K, Ca and Mg. *Fusarium* spp was the major root rot pathogen isolated with high population of up to 14,000 CFU/g. Root rots intensity and seedling infection were high in agro ecological zone LM4 than in agro ecological zone LM1 and varieties KK8 and certified GLP2 had low disease intensity. Farm saved and seeds from the market had high root rot intensity levels. The results showed that poor soil fertility and build-up of soil borne inoculum contributed to high root rots intensity and reduced plant stand. Use of certified seeds and tolerant varieties is effective in reducing the intensity of root rot diseases.

Key words: Common beans, minerals, root rots, soil borne pathogens.

3.2 Introduction

Common bean (*Phaseolus vulgalis*) is an important grain legume used as food security crop and income generation in Kenya (Ochilo, 2013; FAO, 2015). The per capita consumption is projected at 66 Kg per year especially in Western parts of Kenya (Sibiki, 2012; Namugwanya *et al.,* 2014; Petry *et al.,* 2015). Nearly 50% of bean producers sell the produce to urban areas while the canning type is exported to neighbouring countries (Katungi *et al*., 2010; Oshone *et al*., 2014). Production of common bean significantly dropped in the past 20 years largely because the crop is grown by small scale farmers who carry out continuous legume cropping due to decreased land for bean production which led to decline in soil fertility and build-up of soil borne pathogen inoculum (Seremi 2011; Thies, 2016). This has resulted in increased severity of root rot and hence low yields of common beans (Otsyula and Ajang, 1998; Katungi, 2010).

One of the constraints to bean production in East Africa is low soil fertility which causes low productivity (Joseph and Issahaku, 2015; McCann, 2000). According to Beebe *et al*. (2013) and Tairo and Ndakidemi (2013), soils in most common bean growing regions of Kenya have low levels of nitrogen and phosphorus. Studies by Beebe *et al.* (2013) and Argaw (2015) showed that 60% of the bean production areas in Eastern Africa were affected by nitrogen deficiency that led to yield losses of up to 40%. Soil fertility was found to be the major constraint in bean production in Western Kenya (Okalebo *et al.,* 2005; Namugwanja *et al.,* 2014). Proliferation of root rot pathogens is favoured by low soil fertility especially Nitrogen and Phosphorus and soil compaction (Namayanja *et al*., 2010). Studies by Naseri (2014) indicated that organic matter negatively correlated with *Fusarium* root rots.

Root rots in common beans are known to cause significant yield losses of up to 100% (Mwangombe *et al.,* 2008; Mukankuzi *et al.,* 2011; Hergerty *et al.,* 2015). The root rot in common beans is caused by individual or multiple infections of bean plant by *Fusarium* spp, *Macrophomina* spp, *Rhizoctonia* spp and *Pythium* spp (Botelho *et al*., 2013; Mohammed *et al.,* 2013). When they occur as a complex, these causal agents can cause devastating effects on the crop at seedling stage resulting to 100% yield loss under favourable environment (Schwartz, 2011). The disease is difficult to manage as it has various forms of survival and continuous availability of the host due to continuous bean cultivation (Otysula, 2003; Hagerty, 2013). Higher root rot pathogen inoculum in soil is directly linked to increased incidence and severity of root rot of beans (Bhatti and Craft, 2006; Shin- yi, 2012). Therefore this study aimed at determining the effect of soil borne pathogen inoculum on the severity of root rot in common bean.

3.3 Materials and Methods

3.3.1 Description of the study area

The experiment was conducted in Butula and Busire sub-locations, Busia County under Lower agro ecological zone LM1 and Arongo and Rachar sub-locations, Bondo in Siaya County under AEZ LM4 during the long rain cropping season of 2015 (Appendix I and II). Busia is located in Busia County, Western Kenya between latitude 0ºand 0°45′ North and longitude 33°54′ and 34°25′ East (Rutto *et al.,* 2013) and lies at an altitude of between 1216 and 1600 metres above sea level. The area is humid with an average temperature of 23 ºC with a minimum and maximum temperature of 16.2 ºC and 28.7 ºC, respectively. The area receives an average annual rainfall of 1500 mm. Bondo lies at a latitude of 0º14 and 0º 19 North and a longitude of 34º 16’ and 34º 10’ East. The altitude ranges from 1135 to 1200 metres above sea level. Bondo has a modified

equatorial climate with strong influence from the local expansive Lake Victoria, which influences rainfall amounts and distribution (Jaetzold *et al.,* 2009). The area has warm, dry and humid climate with mean annual rainfall between 800 and 1600 mm.

3.3.2 Experimental design, treatments and production of common bean

Common bean varieties from different sources and with varying levels of tolerance to fungal diseases were planted in four experimental sites within two agro ecological zones, LM1 and LM4 in Western Kenya. The varieties were KATX56, KATX69, KK8, and GLP2 with planting seeds sourced from different sources including certified seed from agro dealers, farm saved and seed from the local market. The choice of GLP2 bean variety was based on results from an earlier survey that found it to be the predominant bean variety grown by farmers in western Kenya.

The treatments comprised of KATX56 and GLP2 sourced from the local market, KATX69 and GLP2 sourced from farmers' own saved seed from previous season and certified KK8 and GLP2 from agro dealers. The treatments were laid in a 5M by 5M square plot size with a one metre guard row left around the plot. The crop was planted at a spacing of 30 cm by 15 cm where two seeds were planted in each hill. Each of the six treatments was replicated three times in a Completely Randomized Block Design (CRBD). Weeding was done as recommended at seedling and vegetative stages. Data collected were emergence, stand count, root rot disease distribution, incidence and severity and seedling infection with root rot.

3.3.3 Collection of soil samples and determination of soil nutrient content

Soil samples were collected before planting and the population of soil borne pathogens and nutrient status determined. One kilogram soil sample was extracted from each plot using a soil auger at a

depth of 15-20 cm and mixed to obtain six kilograms composite sample per block as described by Okechukwu and Ekpo (2008). The composite sample was thoroughly mixed, air-dried at ambient temperature (23 ± 2 ºC) and sieved to remove stones and plant debris. A 0.5 kg sub-sample was packed in a khaki bag and labelled according to site and plot and placed in a polythene bag to preserve moisture and kept at 4 ºC before laboratory analysis.

Soil nutrient analysis was carried out at KALRO-NARL laboratories to determine soil organic carbon using calorimetric method described by Anderson and Ingram (1993) while total N was determined by Kjekdahl method described by Hinga *et al*. (1980) and Page *et al*. (1980). Available P was determined by Olsen method (Olsen *et al.,* 1954), while available nutrients (K, Ca, Mg and Mn) and trace elements (Fe, Zn and Cu) were extracted using Mehlich double acid method as described by Mehlich *et al.* (1962). Soil pH and electrical conductivity was determined in a 1:1 (w/v) soil-water suspension with a pH meter.

3.3.4 Determination of population of soil borne fungal pathogens

Soil sub-samples were obtained from the composite samples described in Section 3.3.3. Serial dilutions were made by diluting 1g in 10ml sterile distilled water up to 10^{-3}. One millilitre aliquots of 10^{-2} and 10^{-3} dilutions were used to isolate soil borne fungal pathogens using pour plate method. One milllilitre of each dilution was pipetted into sterile Petri dishes and 20ml of sterile molten PDA media amended with 50ppm streptomycin and 40ppm tetracycline was then added. The content was gently swirled and allowed to settle and solidify as described by Negron-Ortiz (2013).

The isolations were replicated three times and then incubated at room temperature (23 ± 2 ºC) for 7 days. Fungal colonies with similar cultural characteristics were counted and total number of colony forming units per gram of soil calculated as follows:

$$\text{CFU/g} = \text{Total number of colonies} \times \text{Dilution factor}$$

Sub culturing of a representative isolate of each type of the fungal pathogens was done on PDA and SNA and incubated for seven days at room temperature (23 ± 2 ºC) to obtain pure cultures. The fungal isolates were identified by morphological and cultural features such as colony color and type of growth supplemented with microscopic identification using identification keys (Watanabe, 2010).

3.3.5 Isolation of root rots pathogens from stem bases

Ten symptomatic and asymptomatic bean seedlings were randomly collected per plot through destructive sampling in Busia County and Bondo sub-county. The samples were packed in labelled khaki bags and transported to laboratory in a cool box. The stem bases were washed in running tap water to remove soil particles and blot dried. Each seedling stem base was cut into five portions which were surface sterilized in 1.3% sodium hypochlorite for three minutes and rinsed in three changes of sterile distilled water before being blot dried. Five pieces were then aseptically plated in each Petri dish containing PDA amended with 50 ppm streptomycin and 40 ppm tetracycline, replicated three times and incubated for 7-14 days at room temperature. Data was taken on the number of fungal colonies and colony types. Sub culturing of representative individual colonies on sterile PDA and SNA for *Fusarium* spp was done to obtain pure cultures. Identification of the fungi was done based on morphological characteristics such as hyphal septation, conidia shape, size and cultural features such as pigmentation, color of the colonies and type of growth (Watanabe, 2010).

3.3.6 Field assessment of root rot

Assessment of root rot complex was carried out based on symptoms such as chlorosis, stunted growth, wilting, brown discolouration on roots and dark brown lesions on roots (Buruchara *et al.,* 2015). Disease distribution was assessed after every two weeks from the onset of appearance of disease symptoms to the eighth week using a modified scale of 0-2 as described by (Abebe *et al.,* 2014):

Where 0 = no disease, 1 = disease occurs in localized spots, 2 = disease distributed in whole field.

Root rot incidence was calculated as percent number of plants showing symptoms per plot as follows;

$$\text{Root rot incidence} = \frac{\text{Number of plants with root rot}}{\text{Total number of plants per plot} \times 100}$$

Disease severity was determined by assessing the portion of hypocotyls showing root rot based on a scale of 0-3, where 0 = healthy plants, 1 = Mild infection, 2 = Moderately severe, 3 = The plant is severely necrotic and at least half or more of the plants stunted (Buruchara *et al*., 2015; Lithourgidis *et al.,* 2004).

Total disease index was calculated by summing up the scores of diseases distribution, incidence and severity using the formula below (Mc Kinney, 1923)

$$\text{Total disease index} = \frac{\text{Distribution score} + \text{incidence score} + \text{severity score}}{\text{Sum of maximum numerical score}} \times 100$$

Root rot was assessed at two weeks interval from the onset of disease symptoms to the eighth week after emergence.

3.3.4 Data analysis

All data were subjected to Analysis of Variance (ANOVA) using GENSTAT statistical program 12th edition and means obtained were separated using Fishers protected Least Significant difference (LSD) at 5% level of significance.

3.4 Results

3.4.1 Nutrient status of collected soil samples

There was significant variation ($P \leq 0.05$) in soil pH, available phosphorus, potassium, calcium, magnesium, and soil micro elements including copper, iron, zinc and sodium across sites and AEZs (Table 3.1). Sites within LM4 had a near neutral soil pH while soils from LM1 were acidic. Soil nitrogen and soil carbon did not vary significantly among the sites. However, Soils from LM4 were high in phosphorus, potassium, calcium, magnesium and trace elements of copper, iron, zinc and sodium compared to soils from LM1 while manganese did not vary in the two AEZs. Phosphorus levels were high up to 90 ppm in soils from Rachar while low (10 ppm) in soil from Busire. Soil samples from Butula were lower in phosphorus, potassium, calcium and magnesium compared to other sites.

Table 3. 1: Levels of nutrients (ppm and %) in soils sampled from agro ecological zones LM1 and LM4 of Western Kenya

AEZ	Site	pH	N	C	P	K	Ca	Mg	Mn	Cu	Fe	Zn	Na
LM4	Arongo	5.97b	0.10a	0.96a	35.0b	1.52a	14.8a	4.87a	0.73a	5.62c	84.9a	3.5b	0.9a
LM4	Rachar	6.26a	0.11a	1.01a	90.0a	1.52a	14.6a	4.74a	0.74a	7.98b	90.4a	4.7a	0.9a
LM1	Busire	5.09c	0.08a	0.68a	10.0c	0.36b	1.6b	2.19c	0.75a	11.5a	27.4c	2.9c	0.2b
LM1	Butula	4.92c	0.10a	0.98a	31.67b	0.18c	0.8b	0.64c	0.61a	3.67c	41.6b	2.2d	0.1c
	Mean	5.56	0.09	0.91	41.7	0.9	7.96	3.11	0.71	7.19	61.1	3.3	0.5
	LSD ($P \leq 0.05$)	0.22	0.06	0.77	15.13	0.15	1.68	0.73	0.46	1.99	5.5	0.6	0.1
	CV (%)	2.1	31.5	44.8	19.3	8.7	11.2	12.5	34.7	14.7	4.8	8.9	10.2

Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD - Least significant difference at $P \leq 0.05$; CV-coefficient of variation.

3.4.2 Population of root rot pathogens in soil samples

The root rot fungal pathogens commonly isolated from soils sampled from Busia County and Bondo Sub-County were *Fusarium, Pythium, Macrophomina, Rhizoctonia* and *Sclerotinia* (Figure3.1). Tthe structures observed during microscopic identification were conidia for *Fusarium* and *Alternaria* spp and *Pythium* sporangia (figure 3.2).

Figure 3.1: Cultures of root rot causing pathogens isolated from soils and stem bases of symptomatic and asymptomatic bean seedlings**.**

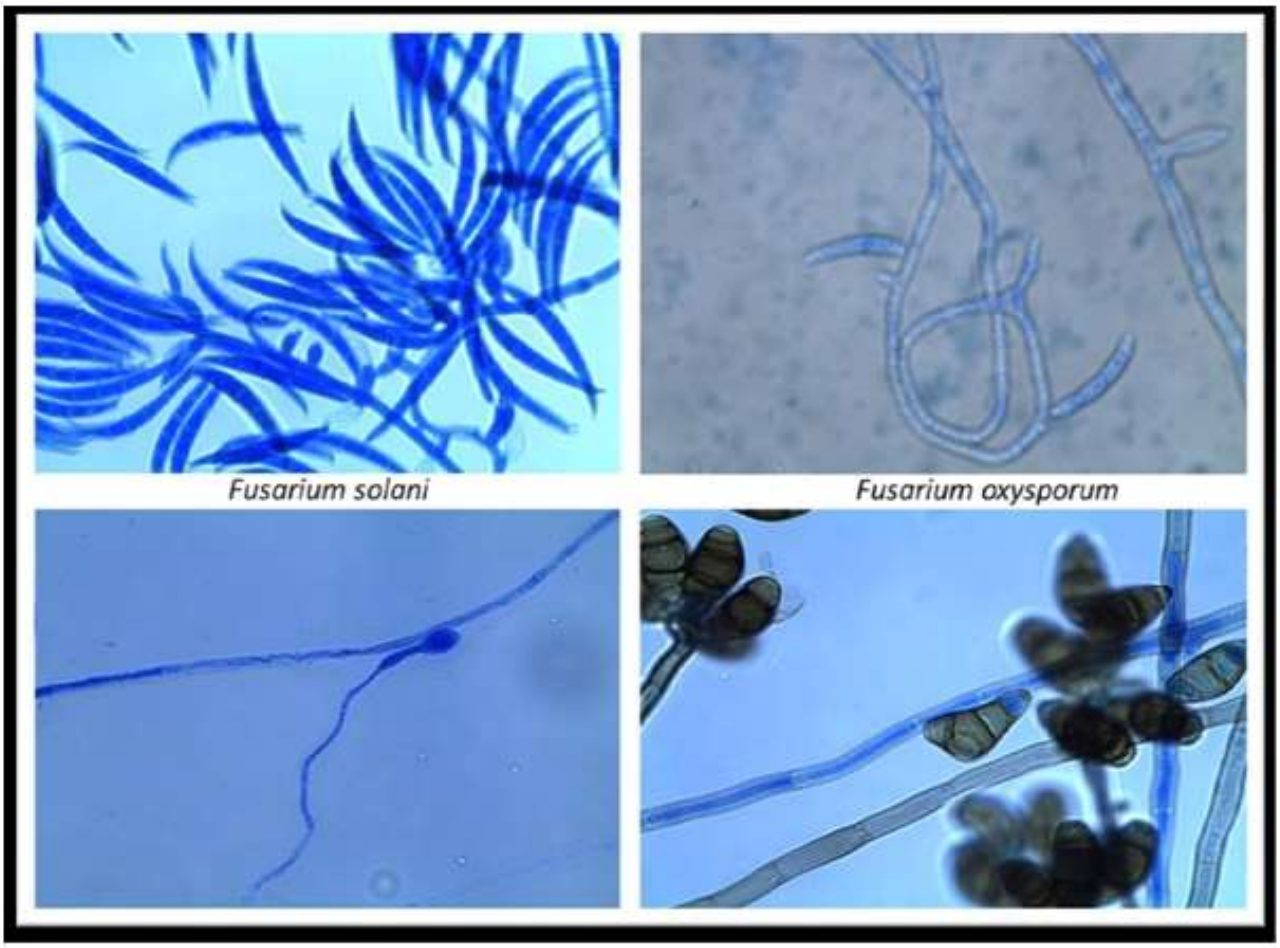

Figure 3.2: Morphological features of root rot pathogens isolated from bean stem bases collected in Busia County and Bondo sub-county during the long rain season of 2015 as seed under the microscope.

All soil samples from both AEZs had high incidence of root rot pathogens. The root rot pathogens isolated were *Fusarium solani, F. oxysporum, Rhizoctonia solani, Pythium ultimum* and *Macrophomina phaseolina*. There were significant differences ($P \leq 0.05$) in the population and incidence of root rot pathogens among the sites (Tables 3.2; Table 3.3). Soil samples from LM1 had a higher population and incidence of *F. solani F. oxysporum* and *Macrophomina* compared to samples from LM4 whereas there was higher population and incidence of *Pythium ultimum* in soil samples from LM4 with a 10% higher incidence of *Pythium* compared to LM1. However, there was no variation ($P \geq 0.05$) in the population and incidence of *Rhizoctonia* between the two AEZs.

Table 3. 2: Incidence (%) of root rot pathogens in soils sampled from four sites within two AEZs in Western Kenya

AEZ	Site	*F. solani*	*F. oxysporum*	*Macrophomina*	*Rhizoctonia*	*Pythium*
LM4	Arongo	47.3ab	21.5a	4.3a	6.6a	10.3b
LM4	Rachar	28.2b	21.4a	4.7a	6.4a	25.2a
LM1	Busire	35.3ab	22.2a	13.3a	6.7a	8.0b
LM1	Butula	50.7a	13.2a	10.1a	9.7a	4.6b
	Mean	40.4	19.6	8.1	7.3	12
	LSD ($p \leq 0.05$)	18.4	10.1	5.7	8.2	11.6
	CV (%)	24.2	27.4	53.5	59.7	51.2

LM1- Lower midland zone one, LM4- Lower midland zone 4; Agro ecological zones. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CVcoefficient of variation.

Table 3. 3: Population (CFU/g) of bean root rot pathogens in soil sampled from four sites within two AEZs of western Kenya

AEZ	Site	*F. solani*	*F. oxysporum*	*Macrophomina*	*Rhizoctonia*	*Pythium*
LM4	Arongo	5,443ab	2,500ab	278a	833a	1,056a
LM4	Rachar	3,710b	1,778b	500a	667a	3,722a
LM1	Busire	4,113b	3,167ab	1,056a	556a	778b
LM1	Butula	9,057a	3,944a	644a	944a	556b
	Mean	5581	2,847	619	750	1,528
	LSD ($P \leq 0.05$)	3,778	1,821	1,251	950	933
	CV (%)	36	34	107	67	32

LM1- lower midland zone one, LM4- lower midland zone 4; Agro ecological zones. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CVcoefficient of variation.

Fusarium solani and *F. oxysporum* were isolated in the highest and lowest incidence with corresponding population in soil samples from Butula and Rachar, respectively. *Macrophomina* was isolated in the highest incidence and population in Busire compared to the other three sites of Arongo, Busire and Butula. *Rhizoctonia* was isolated in the highest incidence and population in Butula but its population was lower in samples from Rachar. *Pythium* spp was isolated in the highest incidence and population in samples from Rachar and Arongo while its population was lowest in samples from Butula and Busire.

3.4.3 Plant stand and intensity of root rots

Percent plant stand varied significantly ($P \leq 0.05$) between AEZs, sites and among the various seed sources. Stand count was 50% higher in LM1 than in LM4 at six and eight weeks after seedling emergence (Tables 3.4; Table 3.5). General reduction in stand count was observed at eighth week compared to sixth week after emergence among all the seed sources (Figure 3.3). Beans grown in Busire had higher plant stand which was 30% higher than in Rachar where the stand count was lowest. Certified seeds of variety KK8 and GLP2 gave the highest stand count of up to 30% higher compared to the farm saved KATX 69 and farm saved GLP2 which had the lowest stand count. Market sourced seeds of variety GLP2 and KATX56 had moderate stand count of above 55%. Similar observations were made at eight weeks after emergence (Table 3.5).

Table 3. 4: Plant stand (%) at six weeks after emergence for four bean varieties from different seed sources in two agro ecological zones

Bean seed source	LM1(Busia)		LM4 (Bondo)	
	Butula	Busire	Arongo	Rachar
KATX56 (Market)	73.6ab	58.0c	58.1a	38.8c
KATX69 (Farm saved)	59.3bc	52.9c	25.3d	28.3d
KK8 (Certified)	84.0a	96.8a	54.2a	54.1b
GLP2 (Certified)	67.6b	97.4a	52.8a	39.9c
GLP2 (Farm saved)	62.3b	54.3c	36.4c	60.8a
GLP2 (Market)	69.9b	71.7b	44.2b	41.0c
Mean	69.5b	71.9a	45.2c	43.8c
LSD ($p \leq 0.05$)	12.4	13.0	8.2	3.3
LSD ($p \leq 0.05$)	Site; 3.6	Treatment; 4.4	Site* Treatment; 8.8	
CV (%)	8.5	8.5	8.7	3.5

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- Rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD- Least significant difference at $P \leq 0.05$; CV- coefficient of variation.

Table 3. 5: Plant stand (%) at eight weeks after emergence for four bean varieties from various seed source in four sites within two agro ecological zones

Bean seed sources	LM1 (Busia)		LM4 (Bondo)	
	Butula	Busire	Arongo	Rachar
KATX56 (Market)	66.7a	53.6c	34.8bc	36.0b
KATX69 (Farm saved)	49.7b	51.0c	30.2c	25.3c
KK8 (Certified)	62.9ab	96.7a	59.7a	57.0a
GLP2 (Certified)	59.1ab	97.2a	48.0ab	37.7b
GLP2 (Farm saved)	61.8ab	52.9c	37.3bc	37.5b
GLP2 (Market)	58.0ab	70.2b	38.2bc	33.5ab
Mean	59.7b	71.0a	41.3c	37.9c
LSD ($p \leq 0.05$)	16.8	12.5	18.3	10.4
LSD ($p \leq 0.05$)	Site; 6.1	Treatment; 7.5	Site* Treatment; 15.0	
CV (%)	13.4	8.4	21.0	13.1

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD- Least significant difference at $P \leq 0.05$; CV-Coefficient of variation.

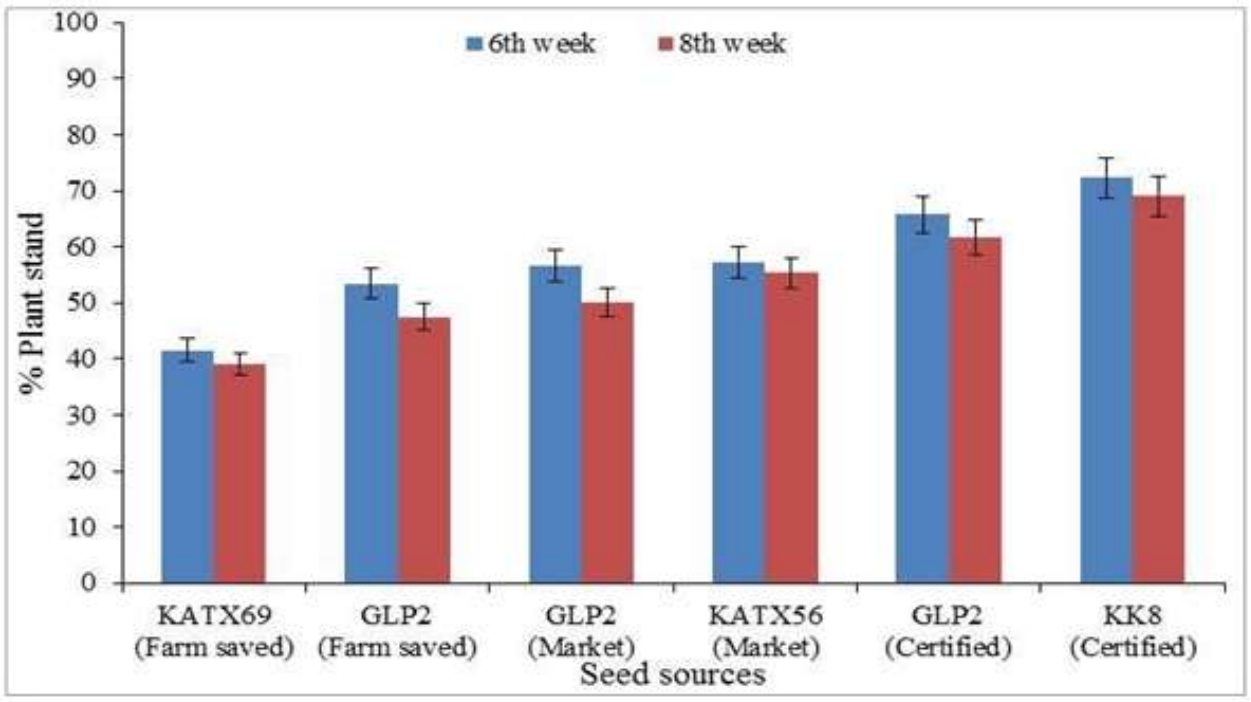

Figure 3.3: Stand count (%) at six and eight weeks post emergence of common beans established from different seed sources at $P \leq 0.05$.

There was variation in root rots intensity between agro ecological zones, sites and among various seed sources. Root rot disease intensity was 10% higher in beans grown in LM4 than in LM1 at six and eight weeks after emergence (Table 3.6; Table 3.7). At six weeks after emergence, bean crops in Arongo and Rachar had high root rot disease intensities whereas at eight weeks, beans in Butula had higher intensities than beans from Arongo and Rachar. Root rots intensity was 20% lower in bean crops in Busire at both assessment periods. Root rot disease intensity also varied with KK8 and certified GLP2 showing lower root rot disease intensities compared to farm saved GLP2, market sourced GLP2 and KATX69 whose intensity was above 57%. However, there was a decline in disease intensity at eighth week for KATX69 and KK8, while no change was observed for KATX56 between the two assessment periods; whereas a steady rise in disease intensity was observed in all GLP2 variety from all sources (Figure 3.4).

Table 3. 6: Root rots intensity at six weeks after emergence for four common bean varieties obtained from different seed sources in two agro ecological zones

Bean seed sources	LM1 (Busia)		LM4 (Bondo)	
	Butula	Busire	Arongo	Rachar
KATX56 (Market)	50.7ab	58.3a	52.1b	71.9ab
KATX69 (Farm saved)	59.2a	57.8a	90.6a	53.7bc
KK8 (Certified)	38.9b	34.4b	42.3b	43.3c
GLP2 (Certified)	57.5a	34.6b	52.2b	53.3bc
GLP2 (Farm saved)	64.2a	36.9b	83.4a	76.8a
GLP2 (Market)	62.9a	52.6a	84.4a	69.8ab
Mean	55.6b	45.8c	67.5a	61.5ab
LSD ($P \leq 0.05$)	15.2	9.8	24.2	18.1
LSD ($P \leq 0.05$)	Site: 7.2	Treatment: 8.9	Site*Treatment: 17.8	
CV (%)	15	11.7	19.7	16.2

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD- Least significant difference at $P \leq 0.05$; CV- coefficient of variation.

Table 3. 7: Percentage root rot intensity at eight weeks after emergence for bean varieties from various seed sources in two agro ecological zones of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)	
	Butula	Busire	Arongo	Rachar
KATX56 (Market)	75.2a	41.2ab	61.5b	56.1b
KATX69 (Farm saved)	83.1a	35.1b	61.9b	62.4b
KK8 (Certified)	37.1b	35.3b	59.9b	48.9b
GLP2 (Certified)	66.7a	35.2b	74ab	67.5ab
GLP2 (Farm saved)	79.4a	48.7a	86.6a	91.3a
GLP2 (Market)	66.7a	35.9b	60.6b	61.1b
Mean	68.0a	38.6b	67.4a	64.6a
LSD ($P \leq 0.05$)	16.3	11.9	17.9	25.6
LSD ($P \leq 0.05$)	Site: 7.5	Treatment: 9.2	Site*Treatment: 18.3	
CV (%)	13.2	17	14.6	21.8

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani X 56 and 69; KK8- Kakamega 8: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD- Least significant difference at $P \leq 0.05$; CV- coefficient of variation.

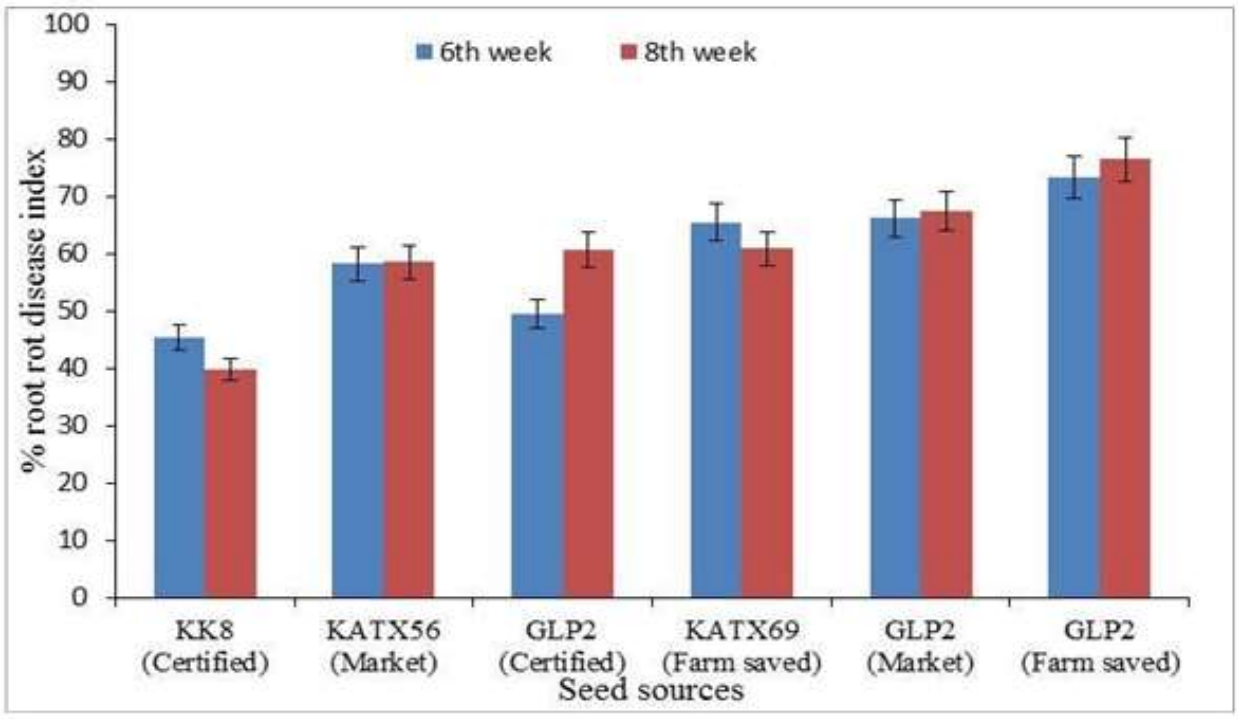

Figure 3.4: Root rots disease intensities (%) at six and eight weeks post emergence of bean seed obtained from different sources at $P \leq 0.05$

3.4.4 Infection of bean stem bases with root rot pathogens

The root rot pathogens isolated from symptomatic and asymptomatic stem bases of the various seed sources were similar to those isolated from the soil. The five pathogens isolated were *Fusarium solani, Fusarium oxysporum, Macrophomina phaseolina, Rhizoctonia solani* and *Phythium* spp. there was no significant difference in the frequencies of the pathogens among the various sites and seed sources. These pathogens were generally isolated in high frequencies in symptomatic than from non-symptomatic stem bases (Tables 3.6; Table 3.7; Table 3.8; Table 3.9).

There were no significant differences in incidence of the root rot pathogens between the two AEZs and among the various seed sources. However, general variations were observed with incidence of *F. solani, Rhizoctonia* and *Pythium* being high on bean stem bases from AEZ LM4 while the incidence of *F. oxysporum* and *Macrophomin*a was high in symptomatic stem bases in AEZ LM1(Table 3.8; Table 3.9). On the other hand, for non-symptomatic stem bases, there was no variation in incidence of *F. oxysporum* and *Macrophomin*a between the two AEZs; although the incidence of *Macrophomina* was 5% higher in AEZ LM4 than in AEZ LM1. Incidence of *Fusarium solani* was highest in both symptomatic and non-symptomatic beans from Arongo and Butula while lower in beans from Busire. *Macrophomina* was isolated in the highest incidence in beans from Busire while *Rhizoctonia* and *Pythium* were high in Arongo and Rachar, respectively. *Fusarium solani* and *F. oxysporum* were isolated in the highest incidence in farm saved GLP2, market sourced GLP2 and KATX69 bean varieties whereas variety KK8 and certified GLP2 had lower incidence but moderate infections were observed for variety KATX56 for both symptomatic and non-symptomatic stem bases.

Table 3. 8: Percentage isolation frequency from stem bases of bean crops from seeds of different sources in agroecological zone LM1 in Busia County

	Symptomatic					Asymptomatic				
Treatments	*F.sol*	*F.oxysp*	*Macrop*	*Rhizoct*	*Pyth*	*F.sol*	*F. oxysp*	*Macrop*	*Rhizoct*	*Pyth*
Butula										
KATX56 (Market)	26.8a	45.2ab	0.0a	4.8a	4.2a	39.3ab	15.0b	0.0a	0.0a	6.7a
KATX69 (Farm saved)	54.8a	40.5ab	0.0a	13.1a	4.8a	35.9ab	33.1abb	8.3a	4.8a	15.0a
KK8 (Certified)	25.4a	28.2b	4.8a	8.5a	0.0a	26.2ab	22.6b	0.0a	0.0a	0.0a
RC (Certified)	23.8a	27.4b	0.0a	0.0a	0.0a	11.1b	20.0b	0.0a	13.3a	24.4a
RC (Farm saved)	54.0a	50.8ab	0.0a	14.3a	0.0a	60.0a	66.7ab	4.2a	0.0a	5.6a
RC (Market)	44.8a	56.6a	0.0a	12.5a	0.0a	63.0a	45.1ab	8.9a	8.9a	6.7a
Mean	38.3	41.4	0.8	8.9	1.5	39.2	33.8	3.6	4.5	9.7
Lsd	28.5	24.3	6.0	22.4	8.0	40.8	33.5	13.0	18.7	22.9
%cv	41.8	33.0	424.6	142.5	300.7	58.5	55.8	204.8	233.2	132.4
Busire										
KATX56 (Market)	24.9a	34.8a	10.8ab	7.4ab	7.4a	52.2a	6.7a	0.0a	0.0a	13.3a
KATX69 (Farm saved)	24.1a	45.2a	30.6ab	0.0b	9.7a	41.3a	16.7a	4.8a	4.8a	0.0a
KK8 (Certified)	20.5a	28.3a	15.7ab	0.0b	0.0a	42.9a	17.3a	0.0a	0.0a	7.9a
RC (Certified)	8.3a	36.7a	4.8b	0.0b	11.8a	38.9a	17.0a	6.7a	6.7a	8.3a
RC (Farm saved)	41.9a	47.2a	19.9ab	10.8ab	7.4a	55.3a	19.8a	5.6a	5.6a	10.8a
RC (Market)	24.4a	44.4a	32.2a	16.8a	0.0a	48.2a	19.2a	4.2a	9.6a	9.6a
Mean	20.0	39.4	19.0	5.8	6.0	46.4	16.1	3.5	4.5	8.3
Lsd	19.4	26.0	24.1	13.0	16.5	33.3	28.1	17.5	13.5	16.1
%cv	45.4	37.1	72.5	125.0	153.0	94.1	34.0	61.1	215.4	108.8

LM1- lower midland zone one; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties: F. sol- *Fusarium solani*; F. oxysp- *Fusarium oxysporum*; Macroph- *Macrophomina phaseolina*; Rhizoct- *Rhizoctonia solani*; Pyth- *Pythium*: Root rot pathogens Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$;LSD- Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

Table 3. 9: Percentage isolation frequency from stem bases of bean crops from seeds of different sources in agroecological zone LM4 in Bondo Sub-county

	Symptomatic					Asymptomatic				
TREAT	F.sol	F.oxys	Macrop	Rhiz	Pyth	F.sol	F. oxys	Macrop	Rhiz	Pyth
Arongo										
KATX56 (MKT)	41.7b	12.5b	0.0b	14.2a	0.0b	26.1b	16.7a	0.0b	3.0a	6.1a
KATX69 (FS)	39.9b	29.4ab	0.0b	8.3a	0.0b	33.3b	39.6a	0.0b	6.7a	3.7a
KK8 (CERT)	15.1b	8.3b	3.3b	0.0a	0.0b	18.0b	15.1a	0.0b	0.0a	0.0a
RC (CERT)	26.0b	28.4ab	5.6b	13.7a	0.0b	23.8b	14.1a	12.2ab	11.6a	6.1a
RC (FS)	75 0a	42.1a	4.8b	21.7a	15.9ab	59.5a	25.4a	5.6b	8.5a	4.8a
RC (MKT)	46.8ab	38.7a	17.0a	18.9a	17.9a	34.6b	34.5a	27.5a	16.7a	21.7a
Mean	40.7	26.6	5.1	12.8	5.6	32.6	24.2	7.5	7.8	7.0
Lsd	30.9	21.2	10.3	24.3	16.6	17.0	29.2	15.6	24.7	19.7
%cv	42.7	44.9	113.7	122.9	165.9	29.4	67.7	116.0	129.0	157.5
Rachar										
KATX56 (Market)	40.7ab	16.7a	0.0a	10.4a	3.3a	23.8bc	17.7a	0.0c	5.6a	8.6a
KATX69 (Farm saved)	39.6ab	36.3a	4.8a	12.5a	0.0a	38.8b	21.4a	3.0bc	12.7a	5.6a
KK8 (Certified)	25.7b	15.0a	12.6a	12.2a	9.0a	18.0c	14.4a	3.3bc	11.1a	7.0a
RC (Certified)	35.9ab	18.2a	7.5a	7.9a	3.3a	15.0c	11.9a	17.6ab	17.6a	8.3a
RC (Farm saved)	55.6a	32.1a	15.3a	0.0a	12.2a	59.5a	38.0a	23.5a	0.0a	17.6a
RC (Market)	25.7b	29.0a	33.3a	6.7a	15.0a	31.1bc	21.7a	3.7bc	10.4a	10.4a
Mean	36.5	24.5	12.3	8.3	7.2	31.0	20.8	8.5	9.6	9.6
Lsd	21.3	20.0	44.4	17.2	14.9	16.1	24.4	7.0	19.7	17.8
%cv	32.9	45.8	203.8	117.3	116.8	29.1	65.8	100.2	115.7	104.5

LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties: F. sol- *Fusarium solani*; F. oxys- *Fusarium oxysporum*; Macroph- *Macrophomina phaseolina*; Rhizoct- *Rhizoctonia solani*; Pyth- *Pythium*: Root rot pathogens Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$;LSD- Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

3.4.5 Correlation among soil nutrients, pathogen inoculum, root rots intensity and infection of bean stem bases

There was negative correlation between soil P and soil Ph. (Table 3.10). The population of soil borne pathogens was negatively correlated to soil carbon, phosphorus and potassium bit positively correlated with soil nitrogen. Stand count had a positive relation with soil nutrients of N, C, P and K but was negatively correlated with the intensity of root rots, the population of soil-borne pathogens and infection on bean stem bases. However, infection on bean stem bases was negatively correlated with soil nutrient levels of N, P, K, C and soil Ph and positively related to intensity of root rot and the population of soil-borne inoculum.

Table 3. 10: Correlation among soil nutrients inoculum levels, stand count, root rots intensity and bean stem base infection

	Soil N	Soil C	Soil P	Soil K	Soil Ph	Root rot intensity	Soil borne	Stand count	Stem base infection
Soil N	-								
Soil C	0.96**	-							
Soil P	0.96**	0.97**	-						
Soil K	0.62*	0.50*	-0.27	-					
Soil pH	0.11	0.37	0.69*	-0.29	-				
root rot Intensity	0.88**	0.97**	0.91**	0.43	-0.56*	-			
Soil borne inoculum	0.97**	0.91**	0.97**	0.80**	-0.51*	0.82**	-		
Stand count	0.87**	0.80**	0.92**	0.92**	0.05	-0.74*	0.97**	-	
Stem base infection	0.86**	0.94**	0.83**	0.17	-0.51*	0.93**	0.72*	-0.55*	-

3.5 Discussion

3.5.1 Nutrient and root rot pathogen inoculum levels in soil

Soil samples from the study sites were below the recommended level for total nitrogen and carbon. While the agro ecological zone LM1, particularly in Butula were below the recommended levels in all the essential elements. The low levels can be attributed to low use of organic and nitrogen fertilizers by farmers, low soil ph, nutrient mining through continuous cropping and poor tillage practices (Wyne *et al.,* 2016). Soil samples from agro ecological zone LM1, particularly in Butula were slightly acidic which agrees with the findings of Van Erp, *et al*. (2015) who reported that the pH range for soils in Busia was between 4.0 and 6.6 and therefore slightly acidic. Kisinyo *et al*. (2014) reported that low soil pH limits the availability of some essential nutrients in soil for example phosphorus is fixed while nitrogen is converted to ammonium or nitrate. Mahdi *et al*. (2005) reported that poor tillage practice reduces soil organic carbon and nitrogen. However, another study by Groffman *et al.* (2009) suggested that both carbon and nitrogen are affected by climate induced moisture and temperature. The high levels of available phosphorus in Busire could probably be due to application of phosphatic fertilizers by farmers and moderate soil pH as shown in the study. This corroborates the findings by Kisinyo *et al.* (2014) and Keino (2015) who reported that use of P, N and organic fertilizers increases the soil available P, total N and total C, respectively in deficient soils with neutral pH.

Variations in level of root rots inoculum between the two AEZs and sites were observed whereby higher populations were detected in soils from AEZ LM1, particularly in Butula where soil pH and fertility were generally low. This agrees with reports by Bardin *et al.* (2004) and Naseri (2014) which stated that root rot pathogens are devastating at moderate soil moisture, hot weather, soil

acidity and poorly fertilized soil similar to conditions in AEZ LM1 hence the high population of root rot pathogens. Högberg *et al.* (2006) also reported that fungal pathogens thrive under low pH and total nitrogen. A study by Namayanja *et al*. (2010) indicated that root rots disease is severe in soils deficient in N, P, AL and Mn toxicity and exchangeable bases which were conditions identified in the current study sites. Soil organic matter provide a microhabitat, substrates and nutrients to the microbial community (Mohammed, 2015) and therefore declining soil fertility reduces microbial population in soil expected to reduce population of soil borne pathogens either through antagonism or competition (Raaijmakers *et al.,* 2008). Agricultural practices such as continuous cultivation, crop rotation and cover cropping also have a strong impact on microbial composition that antagonise on the root rot pathogens (Potthof *et al.,* 2006). Horgberg *et al.* (2006) indicated that microbial community structure is determined by soil pH and C:N ratio and that soil pH negatively correlate with C:N ratio of organic matter and that shift in microbial community is influenced by N availability. Findings by Naseri (2014) also indicated that population of *Fusarium* root rots declined with increase in soil phosphorus. Findings in the current study concur with studies done by Abawi *et al.* (2006) and Mwango'mbe *et al.* (2007) who reported that declining soil fertility due to intensive land use, leads to build-up of root rot pathogens.

Fusarium spp were isolated in the highest incidence and population in all the experimental sites with the highest population being isolated in soil samples from Butula. This agrees with findings by Saremi *et al.* (2011) and Mukanghuzi *et al.* (2011) who reported *Fusarium* spp as the major root rot pathogen of economic importance in legume production. Naseri (2014) further reported that high organic matter results in approximately 50% reduction in *Fusarium* root rots**.** The high population of *Pythium* in LM4 can also be explained by presence of high soil moisture, poor drainage and soil compaction that favour the pathogen development (Isleib, 2014; Binangwe *et*

al., 2016). Rao *et al.* (1978) reported that *Pythium* spp causes severe root rots diseases of corn in poorly drained soil.

3.5.2 Plant stand and intensity of root rots

Root rots disease intensity and plant stand differed significantly between agro ecological zones, sites and among the various seed sources. Root rots intensity was high while stand count was low in beans grown in AEZ LM4 despite low soil borne inoculum level. This could be due to available conditions in LM4 such as flooding, compaction and poor soil properties that favour proliferation of soil borne pathogens as earlier explained resulting to high disease intensity and seedling death and hence reduced plant stand. The disease was more intense in Butula unlike in Busire due to high root rots inoculum in soils and poor soil fertility in Butula resulting in severe root rots infections and reduced stand count. These findings concur with reports by Bardin *et al.* (2004), Abawi *et al.* (2006), Mwango'mbe *et al.* (2007) and Naseri (2014) which stated that root rots are devastating in acidic and poorly fertilized soil and that decline in soil fertility and build-up of root rot pathogen inoculum are caused by intensive land use and continuous legume cultivation.

Root rot intensities were low on bean variety KK8 and KATX56, moderate on bean crop from certified GLP2 and market sourced GLP2 variety but highly intense on bean crop raised from farm saved GLP2 and KATX69 bean varieties. This corroborates a study done by Mwangombe *et al.* (2008), which revealed bean variety GLP2 as a susceptible variety to root rots disease. The fair performance of bean crops raised from market sourced GLP2 and KATX56 bean varieties compared to crop from farm saved seeds, could be due to seed sorting and selection in the market by traders based on visual attributes like seed size and color hence low inoculum on seed (Asfaw *et al.,* 2013). Low root rot infections and intensity on KK8 variety could be due to the tolerance of the variety to root rot infection. Muthomi *et al.,* (2014) reported low incidence of root rots in

tolerant bean varieties KK072, KK15 and KK8 compared to the susceptible GLP2 (Rosecoco) variety. Otsyula *et al.* (2003), Roman-Aviles *et al.* (2004); Batiano *et al.* (2011) and Buruchara *et al.* (2015) attributed the resistant nature of KK8 to root rots to its root architecture hence its adoption in Western Kenya. Seed certification is important in managing root rots in beans, evidenced by high stand count for KK8 and certified GLP2 seed unlike farm saved seed which had low stand count.

The generally high root rot intensities for bean variety GLP2 could be as a result of susceptibility of the variety to root rot infection, high inoculum levels and loss of resistance in farm saved seed due to recycling of seed (Icishahayo *et al.,* 2014). Binagwe *et al.* (2016) also found farm saved seed to be of poor quality due to the presence of survival structures on the surface of the seeds. Recycling of seed can also lead to loss of resistance to fungal infections (Muthii, 2014). Recycling of farm saved and market GLP2 and KATX69 seed and susceptible GLP2 variety to root rot are the reason for their high infection rates.

3.5.3 Infection of bean stem bases with root rot pathogens

The major root rot pathogens isolated from the stem bases were *F. solani, F. oxysporum, Pythium* spp*, Macrophomina* and *Rhizoctonia* spp. with *F. solani* and *F. oxysporum* being the most frequently isolated. *Fusarium* is known to be the major pathogen of the complex causing root rot in common beans (Mukanghuzi *et al.,* 2011).

There was high plant stand and low root rot infections in agro ecological zone LM1 despite the high available inoculum in soil compared to agroecological LM4. Moreover, infections were lower in Busire compared to the other three sites of Rachar, Arongo under agro ecological zone LM4 and Butula under zone LM1. This could be due to soil compaction associated with clay loam soils

in agro ecological zone LM4 and poor soil fertility in Butula. Tu and Tan (1991) working on effects of soil compaction on root rots established that soil compaction increases the incidence and severity of root rots in bean. Scott (1985) and Schwartz *et al.,* (2005) reported low root rots infections on peas in well drained, softer, less dense soil profile and roots are able to penetrate to uninfected soil layer. Compacted soil stresses the plants which react by exuding more metabolites that attract root rot pathogens (Allmaras *et al.,* 1988). Soils from Busire had recommended levels of most essential soil elements which could be the reason for low root rot pathogen population and infections in the respective site. Naseri (2014) reported that organic matter negatively correlated with the population of *Fusarium* root rots. Flooding has been shown to reduce plant stand and increase levels of root rot (Isleib, 2014).

Bean crop from farm saved Farm saved GLP2, market sourced GLP2 and KATX69 had high root rots infection and lower stand count; certified GLP2 had moderate infections and stand count while KK8 and KATX56 had lower root rots infections and higher stand count. This negative relation between root rots infection and stand count for each variety and seed source can be explained by available inoculum in recycled seed causing high root rots severity at seedling stage initiating seedling death and reduced plant population. Work done by Abawi and Ludwig (2010) indicated that bean root rots causes rotting of the root system and seedling death hence reduced plant population. However, contrary observations were made by Valencino *et al*. (2006) who found out that root rot did not reduce plant population in beans. The generally high infections for GLP2 variety regardless of seed source compared to KK8 and KATX56 could be due to susceptible nature of GLP2 variety to infection by root rots. Similar studies done by Mwangombe *et al.* (2008) revealed that GLP2 variety was susceptible to root rots pathogens. Batiano *et al.* (2011) and

Buruchara *et al.* (2015) reported KK8 as a resistant variety to root rot hence its adoption in Western Kenya. In addition the variety could be used as source of resistance in breeding. Recycling of farm saved and market sourced seed lead to build-up of inoculum on the surface and loss of resistance to fungal infections (muthii, 2014). Sabry *et al*. (2013) made similar observation when they isolated *Fusarium solani*, *F. oxysporum* and *Rhizoctonia solani* from bean seed. Recycling of seed and susceptible nature of GLP2 variety to root rot could be the reason for high infection rates on the variety and low stand count.

The current study indicate that low Ph contributed to low soil fertility which consequentially increases soil borne pathogen inoculum levels which in turn increases the intensity of root rot on beans and reduced plant population. The findings further indicates that high soil borne inoculum levels, high incidence of stem base pathogen infection and high intensity of bean root rot on the crop decreased total plant population. However, all the soil nutrients had a positive relation to plant stand. A positive correlation existed among soil borne pathogen inoculum, stem base infection and root rot intensity. This is in agreement with Godoy-Lutz, *et al,* (1996) who reported reduction in stand count and hence yields due to high soil borne inoculum levels causing root rot or damping off and seedlings death. The correlation analysis also indicated that low soil nutrient levels of nitrogen, carbon, phosphorus, Potassium and Ph led to increased population of soilborne pathogens, root rot intensity and stem base infections. These findings corroborates with studies by Naseri (2014) which indicated that soil P negatively correlated with *Fusarium* root rot. Other studies have also shown that diseases in species of the Solanaceae family including *Fusarium* root rot, *Alternaria* blight, *Sclerotium rolfsii* and *Pythium* damping off are increased under low N, P, K and low soil pH conditions (Agrios, 2005). High proportion of P in the soil is in fixed form (unavailable to plants) at low soil pH levels. Phosphorus nutrition improves crop vigour and may

decrease severity of diseases through new growth (Da Silva Ceroz and Fitzsimmons, 2016). K-deficiency increases the concentration of soluble sugars in leaf tissues providing a substrate for many pathogens (Potash Institute, www.ipipotash.org). A study on oil palm revealed that supplying K reduced Fusarium wilt in oil palm (McMahon, 2012). K also supports microorganisms that contribute to biological control in soil borne pathogens (Löbmann *et al.,* 2016). Adequate nutrition helps to mitigate disease damage by replacement of root and shoot tissues and strengthen the crop from attack by less virulent pathogens (Marschner, 1995). Bhatti and Craft (2006) and Shin- yi (2012) reported that root rot pathogen inoculum in soil is directly linked to increased incidence and severity of root rots disease of beans

CHAPTER FOUR
EFFECT OF BEAN SEED QUALITY ON FUNGAL AND BACTERIAL DISEASE PRESSURE OF AND YIELD

4.1 Abstract

Deterioration of quality and build-up of pathogen inoculum in bean seed from informal sources contribute to increased diseases intensity and reduced bean yields. This study was carried out to determine the effect of bean seed quality on disease pressure. Farm saved, market sourced and certified bean seed were evaluated for quality before planting and at harvest. Seed samples were tested for physical purity, germination and bacterial contamination. During crop growth, assessment of foliar diseases was done at sixth and eighth weeks post-emergence. Seed purity, germination and bacterial contamination significantly varied for all seed samples. Except for certified seeds of variety GLP2 and KK8, all seed samples were below the 95% recommended purity level and contained over 15% discoloured and shrivelled seeds. Germination of farm saved variety GLP2 and certified KK8 before planting and at harvest respectively were below the 85% recommended standard. Farm saved and market sourced seeds had high bacterial pathogen inoculum levels of up to 3,187 CFU/seed for *Xanthomonas axonopodis* pv. *phaseoli* and 1,634 CFU/seed for *Pseudomonas savastanoi* pv. *phaseolicola*. Low bacterial pathogen inoculum was detected on certified GLP2 and KK8 varieties. Overall disease intensity for common bacterial blight, angular leaf spot, anthracnose, web blight, *Alternaria* and *Aschochyta* leaf spots varied and was averagely above 50% on crops raised from farm saved and market sourced seeds while yields were below 900Kg/Ha. In contrast, bean crops of certified seeds had below 30% diseases intensity and yield above 1,000 kg/Ha. Poor germination and high bacterial inoculum in farm saved and market sourced seeds contributed to high disease intensity and low yields.

Key words: *Phaseoli vulgaris,* seed contamination, seed transmission, disease intensity.

4.2 Introduction

Good quality bean seed is an important component for improved bean productivity (Rubyogo *et al.,* 2007; Katungi *et al*., 2011). Unavailability and inaccessibility of good quality seeds of improved bean varieties on time and in required quantities are among the major factors contributing to poor bean yields (Endeshaw *et al*., 2010; Gichangi *et al*., 2012). This has prompted many resource-poor farmers to opt for the informal seed supply sources such as own farm saved seed from previous seasons, local markets and community seed exchanges among farmers(Coomes *et al*.,2015; McGuire and Sperling, 2016). Farmers use these channels to replenish their old stock, to expand bean area under production or to obtain new varieties (Maredia *et al*.,1999; Katungi *et al*., 2009; Oshone *et al*., 2014). Formal seed sector only accommodates few cultivars adapted to specific regions due to high cost of producing certified seed (Katungi *et al*., 2011). Formal bean seed supply is expensive costing two to four times the cost of obtaining the seed from the local markets and yield gains do not measure up to the costs of acquiring certified seed (Zerbe, 2001). High cost of disease management and certification standards in formal seed production increases the cost of certified seed (Sperling *et al.,* 1992).

Planting farm saved seed that is untreated and of low quality encourages the spread of seed-borne pathogens resulting in build-up of inoculum which eventually leads to outbreak of disease epidemics causing enormous yield losses (Apole *et al.,* 2003; Icishahayo, 2014; Kadaari, 2015). Damages due to seed-borne pathogens include seed abortion, reduced seed size, shrunken seeds, seed rot, seed necrosis and seed discolouration (Muthii, 2014). Contaminated seed is the primary source of inoculum for seed borne diseases (Fininsa and Tefera, 2010). Seed borne diseases include halo blight (*Pseudomonas savastonoi* pv. *phaseolicola*), common bacterial blight (*Xanthomonas axanopodis* pv. *phaseoli*), bean anthracnose (*Cholletotrichum lindemuthianum*) and viruses (Bean

common mosaic virus and bean yellow mosaic virus). Losses due to common bacterial blight are estimated at 40%, angular leaf spot at 80% and 90% for bean anthracnose in favourable conditions (Mohammed, 2013; Fininsa and Tefera, 2010; Mwang'ombe *et al.,* 2007). There has been need to constantly improve bean seeds through breeding and seed certification programs during seed production and field inspections of seed crop using the International Seed Testing Association (ISTA) standards (Rubyogo *et al.,* 2007, Muthii, 2014). International Seed Testing Association (ISTA, 1999) has set standards for certification of bean seed at 99% varietal purity, 0.95% maximum inert matter, 0.05% maximum other seeds, 85% minimum germination and 14% maximum moisture content. These quality requirements guide seed assessment on the quality status of farmer seed sources (David and Sperling, 1999; Muthii, 2014). This study therefore determined the effect of seed quality on fungal and bacterial disease pressure and productivity of common bean.

4.3 Materials and Methods

4.3.1 Collection of bean seed samples

Seed samples of approximately 500g each of four bean varieties KATX56, KATX69, KK8 and GLP2 were sourced from market, farmers saved seed and certified seed from agro-dealer outlets. At harvest, seed samples of each variety and source were collected from each site. Half a kilogram of each representative sample was packed in a khaki bag, labelled and transported and kept in dry conditions at room temperature awaiting seed quality analysis.

4.3.2 Determination of physical purity of bean seeds

A sub-sample of 50g of each sample replicated three times was used to carry out the purity test following the procedure of ISTA (2013). The three replicates of 50g each of the seed sample were separated into pure seed, inert matter, weed seed and other crop seed. Varietal purity was conducted to separate the main seed variety from shrivelled, discoloured and insect damaged bean seeds and every portion weighed. Separations were done on a white separation board and each fraction placed in a paper bag with a predetermined weight. The weights of individual components were taken using a digital weighing balance. The percentage of each fraction was calculated as follows:

$$\text{Component percentage} = \frac{\text{Weight of each component fraction}}{\text{Total test sample weight (50g)}} \times 100$$

4.3.3 Determination of germination and seedling infection

Germination percentage (GP) of the seeds was determined following the procedures by the International Seed Testing Association (ISTA, 2013). This involved random selection of 50 seeds from each sample and these were replicated three times. Seeds were surface sterilized in 1.3% sodium hypochlorite for three minutes and rinsed in three changes of sterile distilled water. The surface sterilised seeds were plated on a three layer sterile paper towels wetted with sterile distilled water making five rows of ten seeds each. Another three layers of paper towels was used to cover the seeds and wetted evenly using sterile distilled water. The paper towel set up was rolled and placed in moist chambers and incubated at room temperature (23 ± 2 ^{o}C) for 7 days in the presence of light and monitored for seed germination. Observations were done after seven days and data was taken on the number of germinated seeds, mouldy seeds, seedlings showing infections, normal seedlings and abnormal seedlings. Germination percentage was calculated according to the ISTA (1999) formula:

$$\% \text{ Germination} = \frac{\text{Germinated seeds}}{\text{Total seeds}} \times 100$$

4.3.4 Determination of seed borne bacterial pathogens in bean seed

Bacterial infection on seed was determined by agar plate method (ISTA, 2007). Saline solution for extracting bacterial pathogens was prepared by dissolving 8.5g sodium chloride (NaCl) in 1000ml distilled water and a few drops of Tween 20 added. The solution was autoclaved for 15 minutes at 121˚C (Gataitis *et al*., 2013). Each seed sample was thoroughly mixed to obtain a composite sample. The number of seeds in 50g of each sample was counted and the thousand seed weight (TSW) calculated as follows:

$$\text{TSW} = \frac{\text{weight of seed (50g)}}{\text{Number of seed in 50g}} \times 1000$$

Fifty grams of each seed sample was suspended overnight for 16-18 h at 5^{o}C in sterile saline plus Tween 20 (0.02%) in sterile conical flasks. The volume of saline used was equivalent to 1.0 x TSW (g). The containers were shaken to obtain a homogenous extract and the extract was subjected to a 10-fold dilution series up to 10^3 by pipetting 1 ml of the extract into 9 ml of sterile saline. Dilutions 10^2 and 10^3 were plated on nutrient agar by pipetting 1ml onto sterile Petri dishes and then adding about 20ml of sterile molten nutrient agar. Once solidified, the plates were incubated at 28° C in an inverted position for 24 to 48 hours. Typical of each of *Xanthomonas axanopodis* pv *phaseoli* and *Pseudomonas savastanoi* pv *phaseolicola* were counted for each dilution. The numbers of colony forming units (CFU) for each pathogen were calculated by as follows:

$$\text{CFU per seed} = \frac{\text{Calculated CFU}}{\text{Number of seed in 50g}}$$

Pure cultures of the resulting bacteria were prepared by sub culturing single colonies on nutrient agar and identification was based on cultural characteristics such as color and texture

4.3.6 Field observation of bean seed samples and determination of yield components

Bean varieties KATX56, KATX69, KK8, and GLP2 from different sources and with varying level of tolerance to fungal and bacterial diseases were planted in two agro ecological zones, LM1 and LM4 of Western Kenya. The treatments which comprised of KATX56 and GLP2 sourced from the local market, KATX69 and GLP2 sourced from farmers' own saved seed from previous season and certified KK8 and GLP2 from agro dealers were laid in a $25m^2$ plot sizes. Two bean seeds were planted in each hill with a spacing of 30 cm by 15 cm. Each of the six treatments was replicated three times using Completely Randomized Block Design (CRBD). Foliar fungal and bacterial diseases assessed in the field were common bacterial blight, angular leaf spot, bean anthracnose, *Aschochyta* and *Alternaria* leaf spots and web blight. Disease distribution, incidence and severity were assessed after every two weeks from appearance of symptoms on all the bean plants in the three inner bean rows. Disease distribution was assessed using a scale of 0-2, while severity was assessed with a diseases scale of 0-3. Percent diseases incidence was calculated as the number of plants showing symptoms in each plot divided by the total number of plats in each plot. Data on disease distribution, incidence and severity for each disease was taken at sixth and eighth week post emergence and disease intensity for each calculated as follows.

$$\text{Total disease index} = \frac{\text{Distribution score} + \text{incidence score} + \text{severity score}}{\text{Sum of maximum numerical score}} \times 100$$

The agronomic parameters recorded were crop stand counts per plot at complete emergence, flowering and maturity dates. Parameters determined at harvesting were number of pods per plant, biomass and seed yield. Ten plants per plot were randomly selected and the number of pods for each plant determined which was expressed as the average number of pods per plant. At harvest dry bean crops from each plot was uprooted and shelled separately. Total seed and biomass yield from each plot was separated and each weighed using a weighing balance. The yield per plot was finally extrapolated to yield per hacter (Muthomi *et al.*, 2008).

$$\text{Grain yield per hacter} = \frac{\text{Grain yield per plot}}{\text{effective harvested area m2}} \times 10000\text{m2}$$

4.3.5 Isolation of foliar fungal and bacterial pathogens

Isolation of bacterial pathogens from diseased leaves was done using a procedure described by Osdaghi *et al.* (2009). The process involved preparation of bacterial macerates by cutting small sections of symptomatic leave samples from advancing margins of lesion using a sterile scalpel. The chopped tissues were surface sterilised in 1.3% sodium hypochlorite solution for 2-3 minutes and rinsed in three changes of sterile distilled water. The tissues were then macerated with a glass rod in a universal bottle containing 0.5ml sterile distilled water. A sterile wire loop dipped into the suspension was used to make streaks on nutrient agar and incubated for 24-48 hours at 23 ± 2 ^{o}C. The colonies were sub cultured on nutrient agar to obtain pure cultures.

Foliar fungal pathogens were isolated following a modified procedure by Addi *et al.* (2013). This involved placement of the surface sterilised tissues on PDA media ammended with50 ppm streptomycin and 40 ppm tetracycline antibiotics and incubated at room temperature (23 ± 2 ^{o}C)

for 5-7 days. Pure cultures were obtained by sub culturing of each fungal colony type on PDA media. Identification of each isolate fungi was done based on fungal morphological and cultural characteristics**.**

4.3.8 Data analysis

Both field and laboratory data were subjected to analysis of variance (ANOVA) using GENSTAT version 12 and mean separation done using Fishers protected Least Significant difference (LSD) at 5% level of significance.

4.4 Results

4.4.1 Physical purity of the seed

The physical purity parameters assessed for four common bean varieties KATX56, KK8, KATX69 and GLP2 from different seed sources before planting and after harvest included proportions of pure seed, other bean varieties, discoloured seed, inert matter and shrivelled seed in the total seed samples (Figure 4.1). There was significant variation ($P \leq 0.05$) in purity parameters among the different seed sources before planting. Certified seeds of variety GLP2 and KK8 had the highest proportion of pure seeds and lower proportions of other bean varieties, insect damaged and discoloured seeds. Farm saved GLP2 and KATX69 seeds had lower proportions of pure seeds and higher percentage of other bean varieties, insect damaged and discoloured seeds (Table 4.1). The percentage of shrivelled seeds was high in KATX69 and low in GLP2 seed from the market while high proportions of KATX56 seed variety were insect damaged.

Purity parameters for all seed samples after harvest also varied significantly ($P \leq 0.05$) across sites and among the seed sources (Table 4.2). All seed sources had an overall mean purity of 80% which

was 10% higher in seeds sampled from Rachar compared to purity in seed sourced from Butula and Arongo. The proportion of pure seeds was generally highest in Certified GLP2 seed followed by seed of variety KATX56 and GLP2 sourced from the market. Purity levels for seed of variety KATX69 were low even though farm saved variety GLP2 seeds had the lowest proportions in the three sites. The proportion of other bean varieties in the harvested bean seeds was 5% high in bean seed from Rachar compared to Busire which had the lowest proportion (Table 4.3). Farm-saved variety GLP2 and KATX69 seeds had up to 13% of other bean varieties compared to variety KK8 and certified variety GLP2 seeds which had the lowest proportions. Market sourced variety GLP2 and KATX56 had moderate proportions of other bean varieties compared to other seed sources.

There were variations in proportions of inert matter in the harvested seed for the different seed sources across the four sites. Bean seeds from Arongo and Rachar had up to 11% inert matter while Busire had 2% (Table 4.4). Among the seed sources, varieties KATX69 and KK8 seeds had the highest proportions of inert matter which was lower in certified variety GLP2 and KATX56 seeds. Bean seeds from Butula and Arongo had higher fractions of discolored seeds compared to Busire and Rachar. About 10% of varieties KK8, KATX69 and farm saved GLP2 seeds were discoloured. Five percent of certified variety GLP2, 6% of market sourced KATX56 and GLP2 seeds were discolored (Table 4.5). The proportion of shrivelled seed after harvest varied significantly ($P \leq 0.05$) across sites and seed source. The proportion of shrivelled seeds from agro ecological zone LM1 was twice as high compared to seed from agro ecological zone LM4. More than 2.5% of seed from Butula were shrivelled while only 0.5% of seed from Rachar were shrivelled. Bean varieties certified GLP2, market sourced GLP2 and KK8 seeds had less than 1% of their seeds shrivelled while more than 2% of farm saved varieties GLP2 and KATX69 seed was shrivelled (Table 4.6).

Figure 4. 1: Physical purity parameters for planted and harvested bean seed samples

Table 4.1: Percentage seed purity parameters for four bean varieties from different seed sources before planting

Bean seed sources	Pure seed	Other bean varieties	Inert matter	Discolored seed	Shrivelled seed	Insect damaged seed
KATX56 (Market)	80.9c	3.3c	1.3a	5.4b	2.4b	7.1a
KATX69 (Farm saved)	74.9d	8.9b	1.6a	3.4b	9.3a	2.0b
KK8 (Certified)	96.6a	0.1d	2.7a	0.0c	0.0d	0.0c
GLP2 (Certified)	97.8a	0.0d	1.5a	0.0c	0.0d	0.0c
GLP2 (Farm saved)	70.5d	13.1a	2.3a	9.1a	2.6b	2.4b
GLP2 (Market)	89.8b	2.5cd	2.6a	4.2b	1.5c	0.0c
Mean	85.1	4.6	2.0	3.7	2.6	2.1
LSD ($P \leq 0.05$)	4.6	2.5	1.6	3.0	1.0	1.3
CV (%)	3.0	29.6	45.4	44.4	19.4	35.3

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

Table 4.2: Proportion (%) of pure seed in four bean varieties from different sources after harvest in two agro-ecological zones in Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	84.8b	77b	80.8bc	91.4a	83.6b
KATX69 (Farm saved)	64.4d	66.4c	62.2d	83.0c	69.0d
KK8 (Certified)	68.8c	79.4b	84.8b	86.4bc	79.8c
GLP2 (Certified)	92.4a	91.2a	93.8a	91.2a	92.2a
GLP2 (Farm saved)	57.8e	92.8a	56.0e	74.4d	70.2d
GLP2 (Market)	84.4b	89.0a	84.8b	87.8ab	84.4b
Mean	75.4c	82.6b	76c	85.6a	80.0
LSD ($P \leq 0.05$)	3.4	5.0	5.4	4.2	
LSD ($P \leq 0.05$)	Site: 1.6	Treatment: 2.0	site*treatment: 4.0		
CV (%)	2.5	3.3	3.9	2.7	3.0

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each

column are not significantly different at P ≤ 0.05; LSD - Least significant difference at p ≤ 0.05; CV- coefficient of variation.

Table 4.3: Proportion (%) of other bean varieties in bean seeds of four varieties from different seed sources at harvest from two agro ecological zones in Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	1.6bc	16.0b	3.8b	2.2bc	5.8c
KATX69 (Farm saved)	16.8a	25.8a	10.8a	2.0bc	13.8a
KK8 (Certified)	3.8bc	2.4c	1.6c	1.0cd	2.2e
GLP2 (Certified)	0.8c	2.0c	0.2c	0.0d	0.8f
GLP2 (Farm Saved)	17.2a	3.4c	11.4a	11.6a	10.8b
GLP2 (Market)	5.0b	3.4c	4.4b	3.4b	4.0d
Mean	7.4b	8.8a	5.4c	3.4d	6.2
LSD (P ≤ 0.05)	3.6	4.6	2.2	1.8	
LSD (P ≤ 0.05)	Site: 0.6	Treatment: 0.7	Site*Treatment: 1.4		
CV (%)	25.8	28.7	21.7	29.2	26.9

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at P ≤ 0.05; LSD - Least significant difference at p ≤ 0.05; CV- coefficient of variation.

Table 4.4: Proportion (%) of inert matter in seeds of four bean seed varieties from different sources after harvest from two agro ecological zones in Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	0.4c	1.2bc	5.0cd	3.8ab	2.6bc
KATX69 (Farm saved)	3.8b	2.4a	11.2a	5.2a	5.6a
KK8 (Certified)	9.0a	1.4bc	7.8bc	1.8b	4.8ab
GLP2 (Certified)	0.4c	2.0ab	3.0d	3.6ab	2.2c
GLP2 (Farm Saved)	2.6b	0.6c	10.2ab	5.4a	4.6ab
GLP2 (Market)	2.4b	1.8ab	8.0bc	4.8a	4.2ab
Mean	3.0c	1.6d	7.4a	4.2b	4
LSD (P ≤ 0.05)	1.6	0.8	3.2	2.4	
LSD (P ≤ 0.05)	Site: 0.8	Treatment: 1.0	Site*Treatment: 2.0		
CV (%)	30.6	31.3	24	32.3	29.1

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at P ≤ 0.05; LSD - Least significant difference at P ≤ 0.05; CV- coefficient of variation.

Table 4.5: Proportion (%) of discolored seed in bean seed of four bean varieties from different sources after harvest from two agro ecological zones in Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	9.2cd	9.0b	8.2c	2.6c	5.8de
KATX69 (Farm saved)	13.4b	4.2c	13.4b	7.0b	9.6b
KK8 (Certified)	18.0a	16.2a	6.8c	10.6a	13.0a
GLP2 (Certified)	7.2d	4.8c	3.2d	5.6b	5.2c
GLP2 (Farm Saved)	10.6bc	2.8c	21.8a	8.0b	10.8b
GLP2 (Market)	8.6cd	5.0c	8.2c	5.2b	6.8c
Mean	11.2a	6b	10.2a	6.4b	8.4
LSD ($P \leq 0.05$)	3.0	2.2	2.6	2.6	
LSD ($P \leq 0.05$)	Site: 1.0	Treatment: 1.2	Site*Treatment: 2.2		
CV (%)	15.0	20.5	13.5	21.7	16.3

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

Table 4.6: Proportion (%) of shriveled seed in bean seed of four bean seed varieties from different sources after harvest from two agro ecological zones of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	3.4b	2.2a	2ab	0.0b	1.8b
KATX69 (Farm saved)	2.4b	1.2ab	2.4a	2.4a	2.2ab
KK8 (Certified)	0.6c	0.6bc	1.2abc	0.4b	0.8c
GLP2 (Certified)	0.0c	0.0c	0.0c	0.8b	0.2c
GLP2 (Farm Saved)	10.6a	0.4bc	0.8bc	0.6b	3.2a
GLP2 (Market)	0.0c	0.8bc	1.4abc	0.0b	0.6c
Mean	2.8a	0.8bc	1.2b	0.6c	1.4
LSD ($P \leq 0.05$)	1.4	1.0	1.4	0.8	
LSD ($P \leq 0.05$)	Site: 0.4	Treatment: 0.6	Site*Treatment: 1.0		
CV (%)	25.8	65.8	63.3	64.0	45.7

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- Rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

4.4.2 Germination and seedling infection

Germination on paper towels and seed health tests were done to determine proportions of germinated seeds, normal seedlings, abnormal seedlings, mouldy seeds and seedlings with infections before planting and after harvest (Figure 4.2).

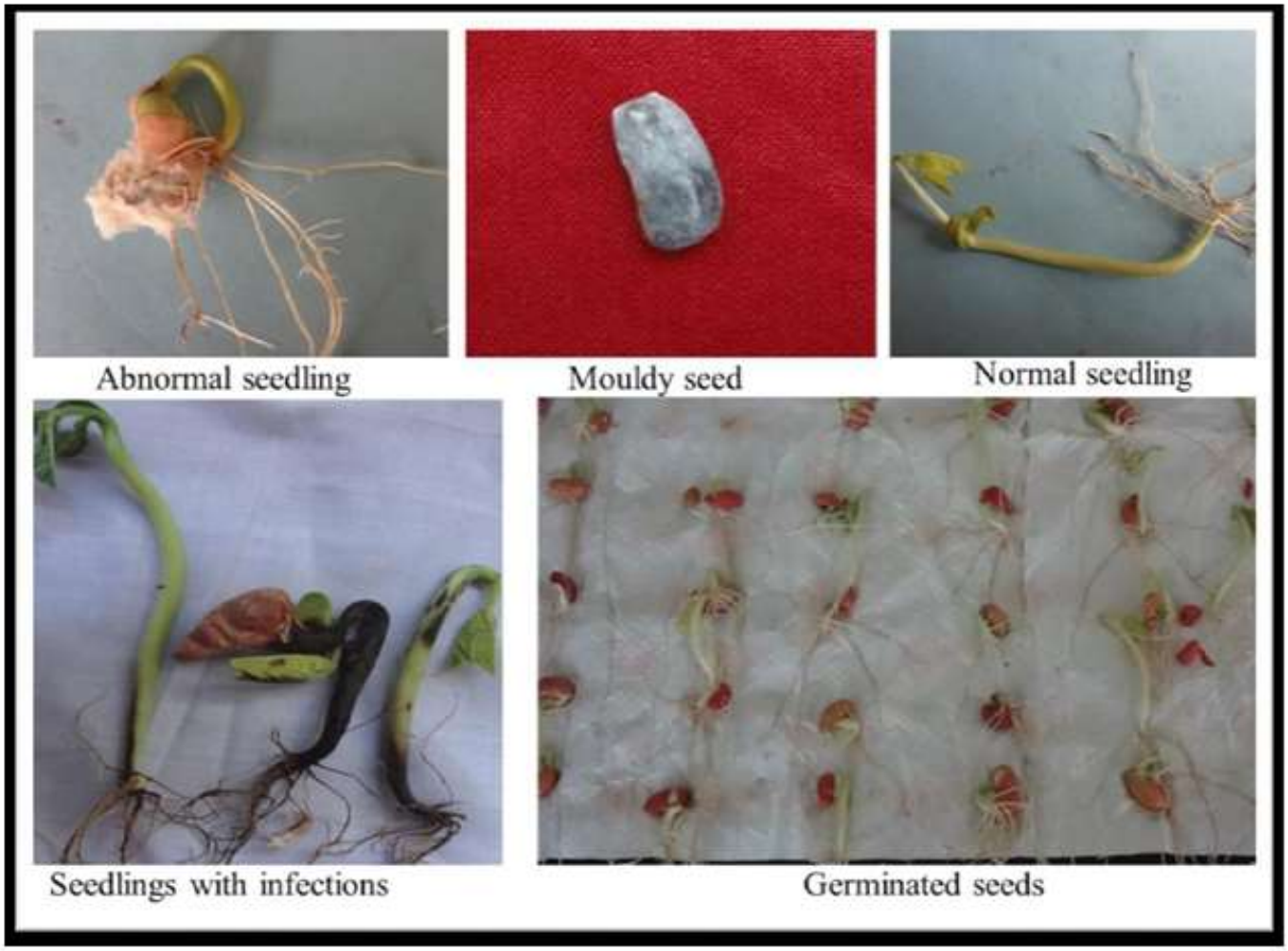

Figure 4. 2: Seed health parameters for bean varieties from different seed sources before planting and at harvest

The mean germination of seeds from different sources before planting varied significantly at $P \leq 0.05$. Seeds of certified varieties GLP2 and KK8 had 100% and 98% germination rate respectively while farm saved variety GLP2 had 76%. Seedlings with abnormalities, infections and mouldy

seeds were low in seed of certified variety GLP2 and KK8 but high in varieties KATX56, KATX69, market and farm sourced GLP2 seeds (Table 4.7).

The proportion of germinated seeds and normal seedlings varied significantly ($P \leq 0.05$) across the sites and seed sources in bean seeds after harvest which was high in bean seed samples from agro ecological zone LM1 unlike LM4. Proportions of germinated seeds were high in bean seed samples from Busire and Rachar and comparatively lower in Arongo (Table 4.8; Table 4.9). Fractions of germinated seeds and normal seedlings were 20% higher in certified variety GLP2 and KATX56 seeds compared to farm saved and market sourced seeds of varieties GLP2 and KATX69. However, the two parameters were lower in KK8 seed samples from Butula. Proportion of abnormal seedlings was 6% lower in seed samples from Busire than Arongo which had the highest. Bean seed of varieties KK8 and KATX69 had 5% more abnormal seedlings compared to certified variety GLP2 which had the lowest (Table 4.10). There was significant variation ($P \leq 0.05$) in the proportion of mouldy seeds and seedlings with infections among the seed sources. The proportion of mouldy seeds was above 22% in farm saved variety GLP2, KK8 and KATX69 seeds and below 20% in certified and market sourced seeds of varieties GLP2 and KATX56. Seedlings with infections were above 48% in varieties GLP2, KK8 and KATX69 and below 44% in certified and market sourced seeds of varieties GLP2 and KATX56 (Table 4.11; Table 4.12).

Table 4.7: Percent seed germination parameters for four bean varieties sampled from different seed sources before planting

Bean seed sources	Germinated seed	Normal seedlings	Abnormal seedlings	Mouldy seeds	Seedlings with infections
KATX56 (Market)	86.0bc	77.3bc	8.7a	7.3b	16.7b
KATX69 (Farm saved)	85.3bc	70.7c	5.3ab	5.3b	8.7bc
KK8 (Certified)	98.7a	96.7a	2.0b	0.0c	0.0c
GLP2 (Certified)	100.0a	99.3a	0.7b	0.0c	0.7c
GLP2 (Farm saved)	76.0c	82.7b	2.7b	22.0a	37.3a
GLP2 (Market)	88.0b	82.7b	5.3ab	4.0bc	18.0b
Mean	89	84.9	4.1	6.4	13.6
LSD ($P \leq 0.05$)	9.7	11.2	4.9	4.3	9.5
CV (%)	6	7.3	65.1	37	38.4

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

Table 4.8: Proportions (%) of germinated seed in bean seed after harvest for bean varieties sampled from different sources in two agro ecological zones in Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	87.2ab	96.6a	68.6bc	97.4a	87.6b
KATX69 (Farm saved)	83.4bc	90.6b	66.6c	92.6ab	83.4bc
KK8 (Certified)	77.4c	96.6a	78.6b	84.6c	84.4bc
GLP2 (Certified)	95.4a	98.0a	96.0a	98.6a	97.0a
GLP2 (Farm Saved)	88.6ab	94.0ab	42.6d	81.4c	76.6c
GLP2 (Market)	88.6ab	94.6ab	66.0c	86.0bc	83.8bc
Mean	86.8c	95.2a	69.8d	90.2b	85.4
LSD ($P \leq 0.05$)	8.4	4	11	7.6	
LSD ($P \leq 0.05$)	Site: 3.0	Treatment: 3.6	Site*Treatment: 7.2		
CV (%)	5.3	2.3	8.7	4.6	5.1

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

Table 4.9: Proportion (%) of normal seedlings for bean varieties sampled from different sources after harvest in two agro ecological zones in Western Kenya

Bean seed sources	LM1(Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	71.4bc	83.4b	51.4b	85.4a	72.8b
KATX69 (Farm saved)	74.6bc	73.4c	42.0b	60.0b	62.6cd
KK8 (Certified)	58.0d	86.6ab	54.0b	60.0b	64.6bcd
GLP2 (Certified)	82.0a	90.0a	80.0a	82.6a	83.6a
GLP2 (Farm Saved)	76ab	84.6ab	24.6c	62.0b	61.8cd
GLP2 (Market)	68.0c	83.4b	46.0b	72.6ab	67.6bc
Mean	71.6b	83.6a	49.6c	70.4b	68.8
LSD ($P \leq 0.05$)	6.4	5.8	14.0	12.4	
LSD ($P \leq 0.05$)	Site: 3.8	Treatment: 4.6	Site*Treatment: 9.2		
CV (%)	4.9	3.9	15.6	9.7	8.2

. LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

Table 4.10: Proportion (%) of abnormal seedlings in bean seed after harvest of bean varieties sampled from different sources in two agro ecological zones in Western Kenya

Bean seed sources	LM1(Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	17.4abc	13.4ab	17.4a	12.0bc	15.0abc
KATX69 (Farm saved)	10.6c	17.4a	24.6a	32.6a	21.4a
KK8 (Certified)	19.4ab	10.0bc	24.6a	36.9a	19.6ab
GLP2 (Certified)	13.4c	8.0c	16.0a	16.0bc	13.4bc
GLP2 (Farm saved)	12.6c	9.4bc	18.0a	19.4b	14.8ab
GLP2 (Market)	20.6a	11.4bc	20.0a	13.4bc	16.4abc
Mean	15.6b	11.6c	20.2a	19.6a	16.8
LSD ($P \leq 0.05$)	6.4	4.4	9.0	10.4	
LSD ($P \leq 0.05$)	Site: 3.2	Treatment: 3.8	Site*Treatment: 7.8		
CV (%)	22.7	21.1	24.7	29.0	28.0

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

Table 4.11: Proportion (%) of moldy seeds in bean seed at harvest for four bean varieties from different sources after harvest in two agro ecological zones in Western Kenya

Bean seed sources	LM (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	19.4b	8.0a	38.0b	2.0cd	16.8ab
KATX69 (Farm saved)	18.0b	10.6a	40.6b	11.4bcd	20.2ab
KK8 (Certified)	30.6a	10.0a	21.4c	26.0a	22.0a
GLP2 (Certified)	9.4c	4.0a	9.4c	0.2cd	6.0c
GLP2 (Farm saved)	8.6c	9.4a	62.0a	19.4ab	24.8a
GLP2 (Market)	19.4b	9.4a	41.4b	13.4bc	20.8ab
Mean	17.6b	8.6c	35.4a	12.2c	18.4
LSD ($P \leq 0.05$)	8.2	7.2	14.8	11.2	
LSD ($P \leq 0.05$)	Site: 3.8	Treatment: 4.6	Site*Treatment: 9.4		
CV (%)	25.9	46.5	23.0	50.0	30.8

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

Table 4.12: Proportion (%) of seedlings with infection in bean varieties sampled from different sources after harvest two agro ecological zones in Western Kenya

Bean seed sources	LM1(Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	78.6ab	14.0c	53.4ab	20.6d	41.6bc
KATX69 (Farm saved)	78.0ab	42.6a	49.4ab	41.4b	50.4ab
KK8 (Certified)	84.0a	33.4b	43.4b	64.6a	56.4a
GLP2 (Certified)	46.6d	15.4c	57.4a	26cd	36.4c
GLP2 (Farm saved)	57.4cd	29.4b	42.6b	63.4a	48.2ab
GLP2 (Market)	79.4ab	14.6c	50ab	33.4bc	44.4bc
Mean	69a	24.8d	49.4b	41.6c	46.2
LSD ($P \leq 0.05$)	12.2	6.2	10.4	11.6	
LSD ($P \leq 0.05$)	Site: 3.8	Treatment: 4.6	Site*Treatment: 9.2		
CV (%)	9.7	13.5	11.7	15.5	12.1

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $P \leq 0.05$; LSD - Least significant difference at $p \leq 0.05$; CV- coefficient of variation.

4.4.3 Bacterial infection of seeds

Xanthomonas axonopdis pv. *phaseoli* (Common bacterial blight) and *Pseudomonas savasatnoi* pv. *phaseolicola* (Halo blight) were isolated in bean seeds before planting and at harvest. Population of *Xanthomonas* was generally high above 2000 CFU/ seed while *Pseudomonas* was as low as 1400 CFU/seed in most of the seed samples. However, at harvest, the population of *Pseudomonas* was high up to 573 CFU/seed in bean seed samples from Busire compared to *Xanthomonas* which was 95 CFU/seed (Figure 4.3Table 13). Before planting, farm saved and market sourced seeds of variety GLP2 and KATX 69 had high population of above 2000 CFU/seed and 900 CFU/seed for *Xanthomona* and *Pseudomonas* respectively while lower population was detected in certified seed of varieties KK8, GLP2 and market sourced seed of variety KATX56 (Figure 4.3). At harvest, higher population for both pathogens was detected in seed samples from agroecological zone LM1 compared to LM4. Population of *Pseudomonas* in agro ecological zone LM1 was twice as high as in agro ecological zone LM4. Bean seed samples from Butula had the highest contamination with both pathogens, while in Busire the population of *Pseudomonas* was 74% higher than that of *Xanthomonas* (Table 4.13). Population of *Xanthomonas* and *Pseudomonas* was high up to 80% and 78% respectively in farm saved seed of variety GLP2 seed compared to certified seed of varieties KK8 and GLP2 which had lower populations.

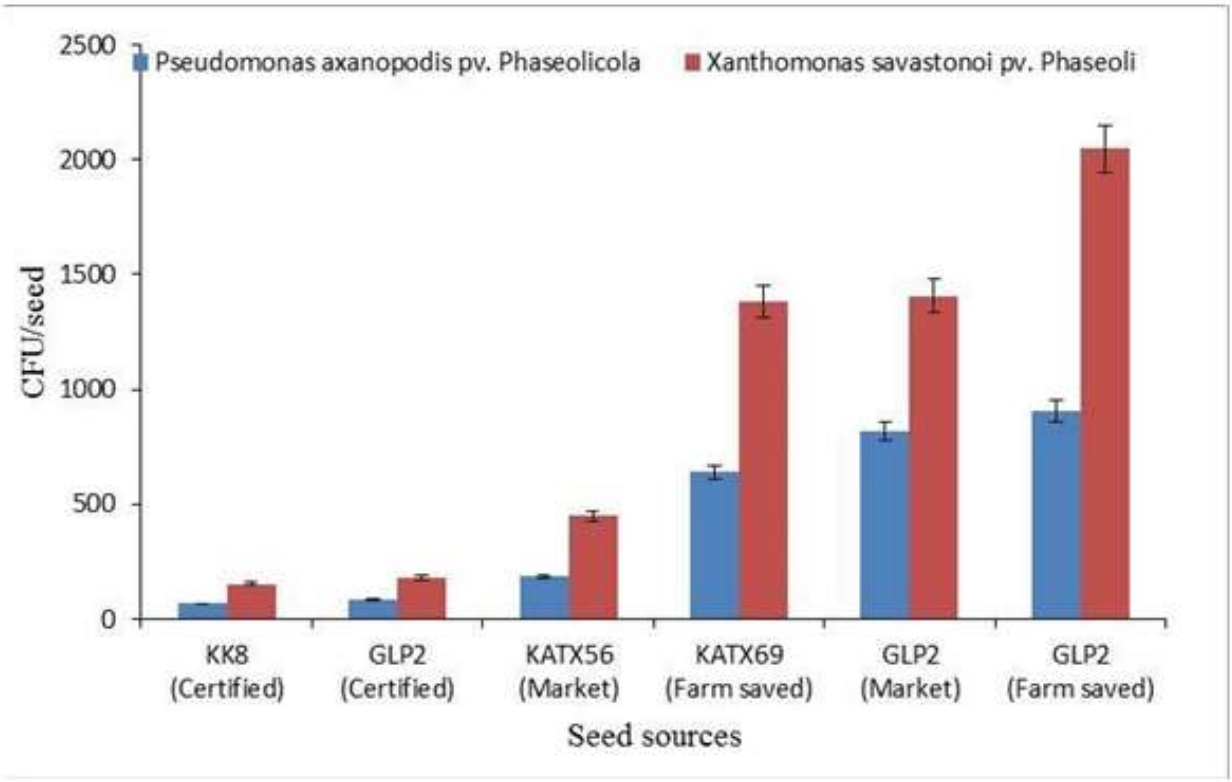

Figure 4. 3 Population (CFU/seed) of *Xanthomonas axanopodis* p.v. *phaseoli* and *Psedomonas savastanoi* p.v. *phaseolicola* in four common bean varieties sampled from different seed sources before planting at $P < 0.05$.

Table 4.13: Population (CFU/seed) of *Xanthomonas axonopodis* pv. *phaseoli* and *Pseudomonas savastanoi* pv. *phaseolicola* detected from bean seeds from different sources after harvest in agro ecological zonesLM1 and LM4 of Western Kenya

Bean seed sources	LM1(Busia)				LM4 (Bondo)					
	Butula		Busire		Arongo		Rachar		Mean	
	Xap	Pseud	Xap	Pseud	Xap	Pseud	Xap	Pseud	Xap	Pseud
KATX69(Farm saved)	1,536b	120d	72a	312a	1,335ab	88b	114b	225b	764ab	186c
KATX56(Market)	1,287b	853bc	82a	785a	1,768a	650ab	113b	32b	812ab	580ab
KK8 (Certified)	118b	40d	77a	436a	337c	252ab	64b	625a	149c	339b
GLP2 (Certified)	523b	215cd	89a	639a	367c	203ab	39b	74b	254c	283bc
GLP2 (Market)	1,517b	923b	60a	602a	844bc	444ab	279a	227b	675b	549ab
GLP2 (Farm saved)	3,187a	1,634a	188a	664a	814bc	1,033a	46b	176b	1059a	877a
Mean	1,361a	631a	95c	573a	911b	445ab	109c	227b	619	469
LSD ($P \leq 0.05$)	1,419	645.6	137.5	530.2	646.2	839.7	113.3	208.6		
LSD ($P \leq 0.05$) Xap	Site: 281.8		Treatment: 245.2				Site*Treatment: 690.3			
LSD ($P \leq 0.05$) Pseud	Site: 240.0		Treatment: 294.0				Site*Treatment: 587.0			
CV (%)	57.3	56.2	79.9	50.9	103.6		57	50.6	67.9	76.2

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; Pseud-*Pseudomonas savastanoi* pv. *Phaseolicola;* Xap- *Xanthomonas axanopodis* pv. *Phaseoli*; bacterial pathogens: KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; GLP2- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $P \leq 0.05$,CV-Coeficientofvariation

4.4.4 Intensity of foliar diseases on beans

Foliar diseases observed in Busia County and Bondo Sub-county included common bacterial blight and angular leaf spot anthracnose, *Aschochyta* leaf spot, web blight and *Alternaria* leaf spot (Figure 4.4). Common Bacterial Blight (CBB) and Angular Leaf Spot (ALS) were the most common foliar diseases affecting bean crop from various seed sources. There were variations in total disease indices for the two diseases in all sites and among the various seed sources. Both common bacterial blight and angular leaf spot diseases had an intensity of above 50% on the bean crops across the four sites. More than 60% intensity of both Common Bacterail Blight and Angular leaf spot on bean crop was observed in Butula. However an intensity of below 50% for the two diseases was observed in Busire and Rachar on the sixth and eighth weeks after emergence (Table 4.14; Table 4.15).

The intensity of CBB was above 70% on bean crop raised from varieties KATX56 and KATX69, moderate below 65% on market sourced and farm saved variety GLP2 while below 53% on certified varieties KK8 and GLP2 crops at sixth week after emergence (Table 4.14). On the eighth week, disease intensity among the seed sources increased from 56 to 60% for market sourced GLP2, farm saved GLP2, KATX56 and KATX69. KK8 and certified GLP2 had lower disease infections of below 43% (Table 4.15).

The intensity for ALS varied significantly ($P \leq 0.05$) across sites and in different seed sources at the sixth and eighth weeks after emergence. Disease intensity for ALS was higher by 20% on bean crop in Butula than Rachar at both assessment times (Table 4.16; Table 4.17). Disease intensity of above 62% was assessed on bean crops raised from farm saved variety GLP2 and KATX69 seed. However, Bean crops raised from market sourced seed varieties KATX56 and GLP2 had moderate

intensities of 54%. On the other hand, disease intensity for ALS was below 46% on bean crop raised from varieties KK8 and certified GLP2 seeds at this period (Table 4.16). On the eighth week post emergence, disease intensity increased by 5% in crops raised from farm saved varieties GLP2 and KATX69 while there was 8% drop in intensity on crops raised from certified varieties GLP2 and KK8 seeds. Bean crops raised from Market sourced seed of varieties KATX56 and GLP2 retained the same intensity levels at both assessment periods (Table 4.17).

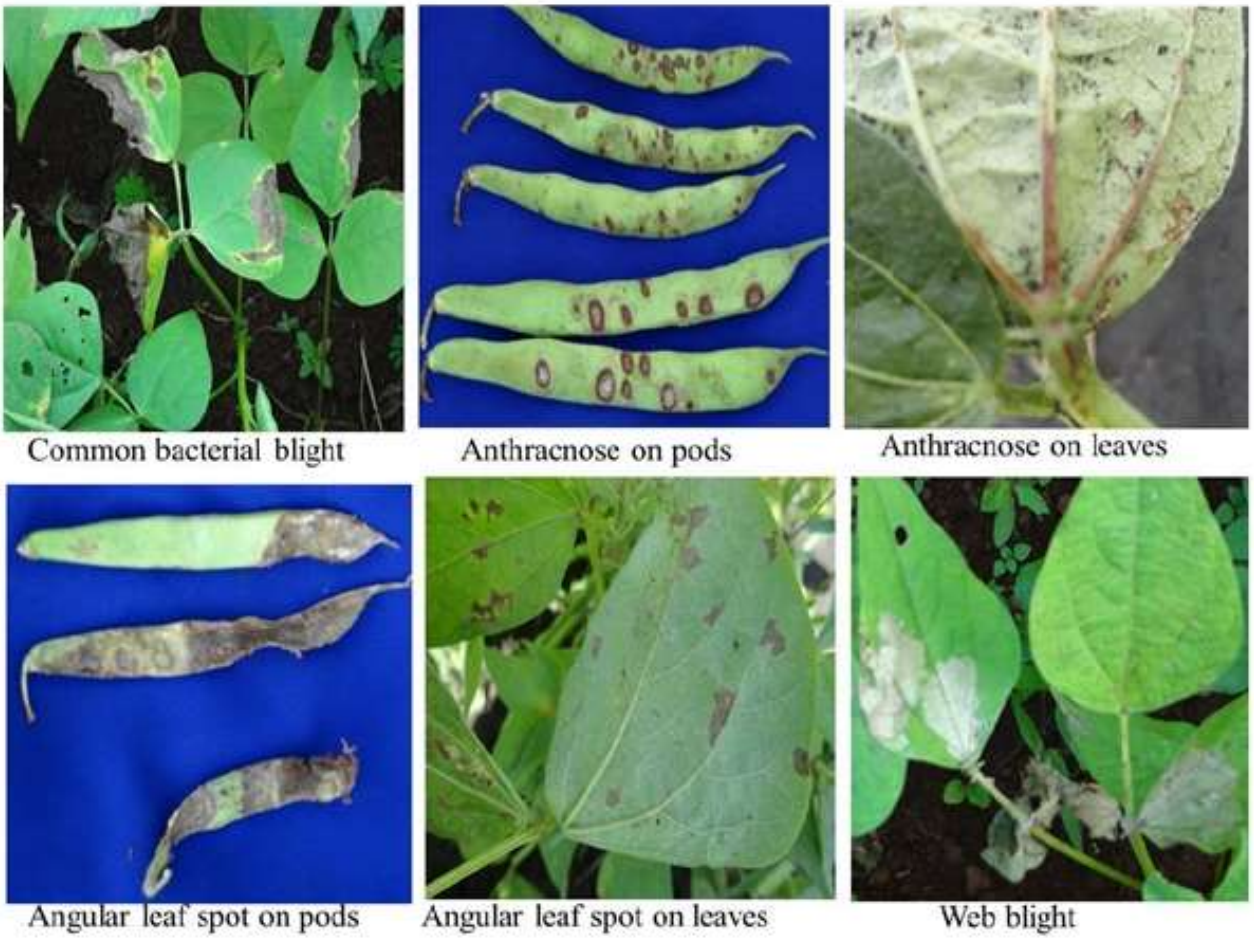

Figure: 4.4 Common foliar diseases of common bean observed in Busia County and Bondo Sub-county during the long rain season of 2015

Table 4.14: Disease intensity (%) for common bacterial blight at six weeks after emergence on four bean varieties from different sources in two AEZs of Western Kenya

Bean seed sources	LM1(Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	74.1b	65.0a	78.6a	77.2a	73.7a
KATX69 (Farm saved)	82.2a	71.1a	79.8a	79.4a	78.1a
KK8 (Certified)	54.2c	11.6c	23.3c	35.8d	31.2d
GLP2 (Certified)	57.7c	37.4b	58.9b	58.2bc	53.1c
GLP2 (Farm saved)	78.1ab	43.9b	75.8a	64.4b	65.6b
GLP2 (Market)	73.0b	53.2ab	76.4a	51.8c	63.6bc
Mean	69.9a	47.0c	65.5ab	61.1b	60.9
LSD ($P \leq 0.05$)	6.3	18.5	15.2	11.1	
LSD ($P \leq 0.05$)	Site: 4.9	Treatment: 6.0	Site*Treatment:12.0		
CV (%)	5	21.6	12.8	10	12

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation.

Table 4.15: Disease intensity (%) for common bacterial blight at eight weeks after emergence for four bean varieties from different sources in AEZs of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	75.2a	57.5a	81.7a	58.9ab	68.3a
KATX69 (Farm saved)	77.2a	50.2a	77.8a	73.3ab	69.6a
KK8 (Certified)	41.2b	11.6b	11.6c	35.2c	24.9c
GLP2 (Certified)	50.0b	43.0a	44.4b	37.0c	43.8b
GLP2 (Farm saved)	78.8a	43.9a	77.8a	77.5a	69.5a
GLP2 (Market)	76.0a	47.6a	70.8a	57.8b	63.1a
Mean	66.4a	42.3c	60.7ab	56.8b	56.5
LSD ($P \leq 0.05$)	16.1	24.3	19.2	18.2	
LSD ($P \leq 0.05$)	Site: 7.7	Treatment: 9.4	Site*Treatment: 18.8		
CV (%0	13.4	31.6	17.4	17.6	20.3

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation.

Table 4.16: Disease intensity (%) for angular leaf spot at six weeks after emergence on four bean varieties from different seed sources in in two AEZs of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	

KATX56 (Market)	74.6ab	62.4a	44.2a	37.8c	54.8bc
KATX69 (Farm saved)	78.7a	47.7abc	55.7a	67.8a	62.4ab
KK8 (Certified)	57.8c	38.5c	43.7a	0.0d	35.0d
GLP2 (Certified)	54.2c	40.0bc	48.9a	44.7bc	46.9c
GLP2 (Farm saved)	77.6ab	48.4abc	67.2a	60.0ab	63.3a
GLP2 (Market)	73.6b	59.3ab	44.8a	38.2c	54.0bc
Mean	69.4a	49.4b	50.7b	41.4c	52.7
LSD ($P \leq 0.05$)	4.1	18.0	27.5	19.7	
LSD ($P \leq 0.05$)	Site: 7.0	Treatment: 8.6	Site*Treatment: 17.2		
CV (%)	3.2	20.0	29.7	26.1	19.9

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation

Table 4.17: Disease intensity (%) for Angular leaf spot at eight weeks after emergence for four bean varieties from different seed sources in in two AEZs of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		
	Butula	Busire	Arongo	Rachar	Mean
KATX56 (Market)	69.2a	49.9bc	56.8ab	38.8b	53.7ab
KATX69 (Farm saved)	79.8a	65.2ab	58.3ab	63.9a	66.8a
KK8 (Certified)	51.7ab	24.5d	23.6c	11.6c	27.8b
GLP2 (Certified)	37.9b	35.9cd	43.1bc	36.8b	38.4b
GLP2 (Farm saved)	78.1a	76.9a	76.1a	51.0ab	70.5a
GLP2 (Market)	57.4ab	71.1ab	51.3ab	46.4ab	56.5ab
Mean	62.3a	53.9ab	51.5b	41.1c	52.3
LSD ($P \leq 0.05$)	27.4	20.7	25.1	24.5	
LSD ($P \leq 0.05$)	Site: 9.9	Treatment: 12.1	Site*Treatment: 24.2		
CV (%)	24.1	21.1	26.8	32.5	28.2

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation

Other foliar diseases assessed were bean anthracnose, web blight, Alternaria and Aschochyta leaf spots. Although there was no significant variation in the intensity of the four diseases general variation in disease intensity was observed. The diseases were generally less severe on bean crop in Busire compared to the other three sites. Bean crops raised from both certified seeds of varieties

KK8 and GLP2 had low disease intensities for the four diseases compared to crops raised from farm saved and market sourced seed of varieties KATX69 and GLP2 (Tables 4.18, Table 4.19, Table 4.20 and Table 4.21). For instance, the intensity of web blight on bean crops of seeds of varieties KK8, KATX56 and certified GLP2 was less than 10% but diseases were above 20% in crop raised from farm saved and market sourced seeds of varieties GLP2 and KATX 69 (Table 4.18). Disease intensities for bean anthracnose, Alternaria and Aschochyta leaf spots were low on varieties KK8 and certified GLP2 crops but were high by 34% on crops from farm saved and market sourced seed of varieties GLP2 and KATX 69. Crops from variety KATX56 had moderate intensities for the four diseases.

Table: 4.18: Disease intensity (%) for web blight at eight weeks after emergence on four bean varieties from different sources in in two AEZs LM1 and LM4 of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	11.bc	11.6a	0.0ab	0.0b	5.8a
KATX69 (Farm saved)	35.3ab	0.0a	35.6a	35.6a	26.6a
KK8 (Certified)	48.2a	30.7a	23.1a	11.6ab	28.4a
GLP2 (Certified)	29.2ab	23.1a	24.4a	23.9ab	25.2a
GLP2 (Farm saved)	34.8ab	17.1a	11.6a	37.2a	25.2a
GLP2 (Market)	0.0c	0.0a	12.2a	23.3ab	8.9a
Mean	26.5a	13.8b	17.8ab	21.9ab	20
LSD ($P \leq 0.05$)	23.8	32.3	33.5	28.1	
LSD ($P \leq 0.05$)	Site: 10.7	Treatment: 13.1	site*treatment: 26.2		
CV (%)	49.3	129.1	103.5	70.4	79.6

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation

Table 4.19: Disease intensity (%) for Alternaria leaf spot at eight weeks after emergence in four bean varieties from different sources in in two AEZs of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		
	Butula	Busire	Arongo	Rachar	Mean
KATX56 (Market)	23.1ab	37.5ab	34.7a	36.5a	32.9a

KATX69 (Farm saved)	23.8ab	0.0c	12.8b	37.7a	18.6a
KK8 (Certified)	0.0b	0.0c	0.0b	0.0c	0.0ab
GLP2 (Certified)	0.0b	0.0c	0.0b	0.0c	0.0ac
GLP2 (Farm saved)	37.4a	23.1b	35.6a	23.7ab	30.0a
GLP2RC (Market)	11.6ab	43.3a	36.4a	23.1ab	28.6a
Mean	16a	17.3a	19.9a	30.2a	18.3
LSD ($P \leq 0.05$)	27.6	18.8	16.7	22.4	
LSD ($P \leq 0.05$)	Site: 7.8	Treatment: 9.5	Site*Treatment: 19.0		
CV (%)	95	59.5	46	61	63

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation

Table 4.20: The disease intensity (%) for bean anthracnose at eight weeks after emergence in bean varieties from different seed sources in in two AEZs of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	11.4b	0.0a	11.7ab	23.3ab	11.6ab
KATX69 (Farm saved)	11.7b	23.1a	12.2ab	24.3ab	17.8ab
KK8 (Certified)	0.0b	0.0a	0.0b	0.0b	0.0b
GLP2(Certified)	0.0b	11.7a	0.0b	0.0b	2.9b
GLP (Farm saved)	37.1a	23.2a	24.4ab	37.8a	30.6a
GLP2 (Market)	34.9a	0.0a	32.9a	30.3a	25ab
Mean	15.8a	9.7ab	13.9a	19.3a	14.7
LSD ($P \leq 0.05$)	22	27.5	28.6	23.4	
LSD ($P \leq 0.05$)	Site: 9.1	Treatment:11.1	Sites*Treatment:22.2		
CV (%)	76.2	156.8	113.4	66.8	92.3

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation.

Table 4.21: Disease intensity (%) for Aschochyta leaf spot at eight weeks after emergence on bean varieties from different sources in in two AEZs of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	

KATX56 (Market)	11.6bc	23.1ab	24.4ab	24.4ab	20.9ab
KATX69 (Farm saved)	41.2a	22.9ab	37.4a	34.7a	34.0a
KK8 (Certified)	0.0c	23.2ab	0.0b	0.0b	5.8b
GLP2 (Certified)	0.0c	0.0b	0.0b	0.0b	0.0b
GLP2 (Farm saved)	35.3ab	0.0b	23.2ab	36.7a	23.8ab
GLP2 (Market)	23.1abc	35.3a	24.3ab	11.7ab	23.6ab
Mean	18.5a	17.4a	18.2a	17.9a	18.0
LSD ($P \leq 0.05$)	23.9	25.6	29.3	23.9	
LSD ($P \leq 0.05$)	Site: 9.0	Treatment: 11.1	Site*Treatment: 22.2		
CV (%)	71.1	80.9	88.4	73.2	74.6

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation

4.2.5 Overall disease intensity for four common bean varieties sourced from different seed sources

There was significant variation ($P \leq 0.05$) in total disease indices for the common bean diseases among the various seed sources and sites at sixth and eighth weeks after emergence. There was 22% increase in disease intensity on the eighth week compared to the sixth week (Table 4.22; Table 4.23). Total disease index was 15% higher on bean crop in Butula than Busire at both assessment periods. Overall disease intensity was 60% on the eighth week on crops raised from farm saved seed of varieties KATX69, GLP2 and KATX56 unlike 40% on the sixth week. However, overall intensity was below 28% and 52% at both sixth and eighth weeks after emergence respectively for variety KK8 and certified GLP2 crops. Moderate disease intensities were observed on market sourced seed variety GLP2 at both periods.

Table 4.22: Total disease index (%) at six weeks after emergence for four bean varieties sourced from different sources in in two AEZ LM1 and LM4 of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	38.9cd	32.5a	36.5ab	36.3b	36.0ab

KATX69 (Farm saved)	49.7ab	26.5ab	42.3a	43.9a	40.6a
KK8 (Certified)	33.5de	19.9b	20.1c	20.1d	23.4bc
GLP2 (Certified)	30.5e	21.9b	32.0b	27.8c	28.0b
GLP2 (Farm saved)	50.7a	27.3ab	43.0a	41.5ab	40.6a
GLP2 (Market)	42.2bc	33.3a	44.3a	35.1b	38.7ab
Mean	40.9a	26.9c	36.0b	34.1b	34.6
LSD ($P \leq 0.05$)	7.5	9.4	9.5	6.6	
LSD ($P \leq 0.05$)	Site: 2.9	Treatment: 3.6	Site*Treatment: 7.2		
CV (%)	10.0	19.1	14.4	10.6	12.6

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation

Table 4.23: Total disease index at eight weeks after emergence for four bean varieties sourced from different seed sources in two AEZs LM1 and LM4 of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	68.6ab	56.7ab	60.5ab	56.4b	60.6ab
KATX69 (Farm saved)	71.4ab	60.1a	59.7ab	74.8a	66.5a
KK8 (Certified)	61.7ab	31.1d	41.1c	37.7c	42.9bc
GLP2 (Certified)	59.0b	40.9cd	50.4bc	57.6b	51.9b
GLP2 (Farm saved)	67.2ab	45.0bc	67.3a	64.8ab	61.1ab
GLP2 (Market)	74.8a	54.5ab	59.6ab	50.6bc	59.9ab
Mean	67.1a	48.1c	56.4b	57.0b	57.2
LSD ($P \leq 0.05$)	12.1	13.0	12.1	13.4	
LSD ($P \leq 0.05$)	Site: 4.9	Treatment: 6.1	Site*Treatment;12.1		
CV (%)	9.9	14.9	11.8	12.9	12.9

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation

4.2.6 Yield and yield components

There were significant differences ($P \leq 0.05$) in seed yield, number of pods per plant and biomass among bean crops planted with bean seeds from different sources across sites within two agro ecological zones. The mean seed yield was below 1000 kg/Ha but generally high above 1000 kg/Ha

in agro ecological zone LM4 unlike LM1 (Table 4.24). However, seed yield was highest in Arongo but lowest in Butula (Table 4.24; Table 4.25). Bean seed yield was above 1100kg/Ha for variety KATX 56 and certified variety GLP2 but was below 700 kg/Ha for farm saved variety GLP2. Market sourced GLP2, KK8 and KATX69 varieties had moderate seed yields ranging between 800 to 1000 kg/Ha (Table 4.24).The number of pods per plant was averagely seven pods per plant which was highest in Arongo with 10 pods per plant and lowest in Butula with three pods per plant (Table 4.25). The average number of pods was above 7 pods per plant for variety KATX56 and certified variety GLP2 but was below 5 pods in farm saved variety GLP2. Biomass yield was high above 1200kg/Ha in Arongo and lowest in Rachar, however there was no biomass data taken in Butula as it had been destroyed by the farmer by the time of data collection (Table 4.26) dry matter was generally high above 1200 kg/Ha for variety KK8 and KATX56 and below 750kg/Ha for farm saved variety GLP2.

Table 4.24: Bean seed yield (kg/Ha) for four bean varieties from different seed sources in in two AEZ LM1 and LM4 of Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	548ab	1,078c	2,411a	1,321a	1,339a
KATX69 (Farm saved)	212c	863d	1,354b	1,083b	878b
KK8 (Certified)	337bc	1,045c	1,394b	1,011b	947b
GLP2 (Certified)	646a	1,622a	1,017bc	1,409a	1,173a
GLP2 (Farm saved)	416b	1,308b	392c	532c	662c
GLP2 (Market)	227c	722e	1,658b	986b	898b
Mean	398c	1,106b	1,371a	1,057b	983
LSD ($P \leq 0.05$))	165.9	134.4	750.7	100.9	
LSD ($P \leq 0.05$)	Site: 139.5	Treatment: 170.9		Site*Treatment: 341.8	
CV (%)	22.9	6.7	30.1	5.2	21.2

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation.

Table 4.25: Average number of pods for four bean varieties from various seed sources in two AEZs in Western Kenya

Bean seed sources	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	3.67a	8.7a	11.3a	8.3a	8.0a
KATX69 (Farm saved)	2.0b	8.3a	9.0b	8.7a	7.0ab
KK8 (Certified)	2.3b	8.7a	8.7b	8.0a	6.9ab
GLP2 (Certified)	2.7ab	7.0b	12.0a	8.7a	7.6ab
GLP2 (Farm Saved)	2.3b	6.0b	7.7b	5.0b	5.3b
GLP2 (Market)	2.7ab	6.3b	8.7b	10.0a	6.9ab
Mean	2.6c	7.5b	9.6a	8.1b	6.9
LSD ($P \leq 0.05$)	1.0	1.3	2.3	2.0	
LSD ($P \leq 0.05$)	Site: 0.6	Treatment: 0.8		Site*Treatment: 1.5	
CV (%)	21.7	9.7	13.2	13.8	13.4

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation.

Table 4.26: Biomass (kg/Ha) for four bean varieties from different sources and in various sites within two AEZs in Western Kenya

Bean varieties	LM1 (Busia)		LM4 (Bondo)		Mean
	Butula	Busire	Arongo	Rachar	
KATX56 (Market)	*	803b	2022a	800a	1,208a
KATX69 (Farm saved)	*	784b	1320b	653a	919b
KK8 (Certified)	*	1,508a	1,457b	787a	1,250a
RC (Certified)	*	751b	1,313b	813a	959ab
RC (Farmer saved)	*	951b	650c	627a	743bc
RC (Market)	*	929b	707c	907a	848b
Mean	*	954b	1245a	764c	988
LSD ($P \leq 0.05$)	*	431.1	372.4	214.7	
LSD ($P \leq 0.05$)	Site: 131.0	Treatment:185.3		Site*Treatment:320.9	
CV (%)	*	31.9	16.4	12.4	19.6

LM1- lower midland zone one; LM4- lower midland zone 4; agro ecological zones; KATX56 and KATX69- Katumani x 56 and 69; KK8- Kakamega 8; RC- rose coco: Bean varieties. Means followed by the same letter(s) in each column are not significantly different at $p \leq 0.05$; LSD- Least significant difference at $p \leq 0.05$; CV-coefficient of variation

4.2.7: Correlation among soil nutrients, diseases, seed health parameters and seed yield

Soil carbon, nitrogen, phosphorus and potassium were positively correlated with total seed yield per hacter (Table 4:27). Both soil carbon and phosphorus were negatively correlated with the intensity of foliar diseases; however, diseases intensity had a negative relation with soil nitrogen. There were significant positive correlation between the number of shrivelled and discoloured seed and the intensity of foliar diseases on the crop. However, negative correlation existed between number of shrivelled and discoloured seed and total seed yield per hacter. The number of seedlings showing infection and the population of bacterial pathogens in seed were also positively correlated with foliar disease intensity but were negatively correlated with total seed yield. The intensity of foliar diseases on bean crop in the experimental sites however had a negative relation with total seed yield per hacter.

Table 4.27: Correlation among soil nutrients, bean diseases and yield components of four common bean varieties from different seed sources in various sites within two agro ecological zones

	Soil C	Soil P	Soil K	Soil N	Foliar disease intensity	Bacterial infection in seed	Infected Seedlings	Shrivelled seed	Discolou red seed	seed yield
Soil C	-									
Soil P	-0.97**	-								
Soil K	0.50*	-0.69*	-							
Soil N	0.96**	-0.96**	0.62*	-						
Foliar disease intensity	-0.76*	-0.59*	-0.18	0.61*	-					
Bacterial infection on seed	0.48	-0.36	-0.30	0.24	0.83**	-				
Infected seedlings	-0.11	-0.58*	-0.16	0.54*	0.97**	0.93**	-			
Shrivelled seed	0.32	-0.12	-0.02	0.09	0.84**	0.92**	0.88**	-		
Discoloured seed	-0.22	-0.47	-0.19	0.36	0.87**	0.99**	0.96**	0.89**	-	
Seed yield	0.59*	0.60*	0.71*	0.73*	-0.76*	-0.57*	-0.67*	-0.83**	-0.55*	-

*- Significant; **- Highly significant at 5% level of probability

4.5 Discussion

4.5.1 Physical purity, germination and bacterial contamination in seed

There were significant variations in the level of physical purity of seed, germination capacity and level of bacterial contamination on seed from the various sources. All seeds sampled before planting and after harvest, had a physical purity of between 56 and 93% which was below the 95% minimum pure seed standard recommended by ISTA (1999). However, certified variety GLP2 seed had higher purity levels compared to farm saved varieties GLP2 and KATX69 which had lower proportions of pure seed and high levels for impurities. This could be attributed to seed certification standards met in the production of certified seed. The findings agree with Boland *et al.* (2001) who reported low fraction of weed and foreign material in certified seed compared to farm saved seed. In addition, Graven *et al.* (2004) and Scott *et al.* (2003) reported that low purity levels in seed could be due to poor crop husbandry and post-harvest management practices by farmers such as threshing, stage of harvesting, drying and storage. Further, certified seeds are genetically pure and free from diseases, physical damage and immature seeds (IRRI, 2013).

The high proportions of discoloured and shrivelled seeds in farm saved seed of varieties GLP2 and KATX69 compared to certified seed lots of varieties GLP2 and KK8 could be attributed to high prevalence of seed borne diseases in recycled seed leading to build- up of inoculum (Icishahayo *et al*., 2009; Osborn *et al*., 2010). These findings agree with studies by Asfaw *et al.* (2013) and Boersma *et al.* (2015) who reported that farm saved seed is of poor quality since seed selection is based on physical quality attributes like color and size and therefore seed in the subsequent seasons become discoloured and shrivelled due to infections. In addition, Dube *et al*. (2014) demonstrated that hand picking of discoloured seeds from farmer retained seeds greatly reduced fungal

infections. However, contrary observations to these were made by Oshone *et al.,* (2014) who reported 98% purity level in bean seed from small scale farmers in Ethiopia.

Seed samples of certified varieties GLP2, KK8 and market sourced variety KATX56 met the minimum recommended germination standards of 85% (ISTA, 1999) while seed samples of farm saved varieties GLP2, market sourced GLP2 and KATX69 had lower than the recommended standard. The low germination rate in variety KATX69, farm saved and market sourced variety GLP2 could be attributed to poor pre and post-harvest handling and storage practices by farmers (Khalequzzaman *et al.,* 2012). Similar findings were obtained by Muthii (2014), Msuya and Stefano (2010) who reported threshing and other post-harvest processes by farmers lowers the germination capacity and seedling vigor. In addition, Osborn *et al.* 2010, Pradhan and Badola (2012) and Shaban (2013) stated that low germination of farmer saved seed is due to long storage periods in poor conditions and high percentage of discoloured seeds and inert materials.

The current study also found that except for certified seed samples of variety GLP2 and KK8 seeds before planting, all other seed samples had above 40% mouldy and infected seedlings. This could be due to high inoculum levels in seed resulting from recycling of bean seed by farmers (Icishahayo *et al*., 2009). Similar studies done on sunflower seeds by Caldeira *et al.* (2015) indicated that infection of seed by fungal pathogens may reduce germination capacity through damage to the seedlings. Poor seed germination and seedling infections lead to low plant population that ultimately result in low bean yield (ISTA, 2015). High inoculum load on seed and lack of seed certification schemes could have been the reasons for high seed rotting in farm saved and market sourced GLP2 and KATX 69 seeds (Icishahayo *et al.,* 2009).

High levels of mouldiness and infected seedlings were observed in agro ecological zone LM4 particularly in Arongo. This can be explained by the humid climatic conditions experienced in the region (Jaetzold *et al*., 2009) and poor post-harvest handling practices by the respective farmer who harvested the crop in wet conditions and poor drying leading to seed rotting. These observations concur with reports by Makelo (2010) and Boersma *et al.* (2015) who reported variation in seedling infections and higher prevalence of bean diseases in some agro-ecological zones due to favourable weather conditions for disease development in a particular zone. High seed rotting in seed samples of variety KK8 was due to the variety being late maturing (KARI, 2008) and it was harvested at the same time with the other early and medium maturing varieties before it fully dried leading to rotting. Healthy seed is therefore the most important agricultural input affecting crop yield (Diaz *et al.,* 1998).

Seed borne fungal and bacterial diseases are a threat to legume production and seed is the main source of inoculum for bacterial diseases (Narayan and Ayodhya, 2013). *Xanthomonas axonopdis* pv. *phaseoli* (Xap) and *Pseudomonas savasatnoi* pv. *phaseolicola* (Psp) were the major bacterial pathogens isolated from seed before planting and at harvest. *Xanthomonas* was the most frequently isolated pathogen of the two with high population in LM1. Western Kenya experiences warm and humid conditions which favour Xap bacteria unlike Psp which is favoured by cool climate (Jaetszold, 2009; Schwartz, 2011; Wohleb, 2011). A study by Karavina *et al.* (2008) further explain the influence of geographical area, climate, cultural practices and bacterial strains on the survival of bacterial pathogens in soil, plant debris and seed hence the availability of inoculum in the next season.

High population of both pathogens were detected in farm saved and market sourced seed of varieties GLP2 and KATX69 before planting and at harvest compared to certified seeds of varieties KK8, GLP2 and KATX 56 sourced from the market which had lower contamination. High frequencies of these pathogens in farm saved seed of varieties GLP2 and KATX69 and market sourced GLP2 could be due to build-up of inoculum in seed as a result of recycling of own saved seed by farmers (Icishahayo *et al.,* 2009). In addition, Karavina *et al.* (2008), Wekesa (2010) and Oshone *et al.* (2014) reported high population of bacterial and fungal contamination in farm saved seed compared to certified seed. Seed is therefore the main source of inoculum for bacterial diseases and consequently a threat to productivity of common bean (Sileshi *et al.,* 2014). This further explains the high prevalence of common bacterial blight in farm saved and market sourced seeds in the two agro ecological zones.

4.5.2 Intensity of fungal and bacterial foliar diseases of beans

Major foliar diseases of common bean were common bacterial blight and angular leaf spot diseases. Other diseases included anthracnose, web blight, Aschochyta and Alternaria leaf spots. The intensity of common bacterial blight and angular leaf spot were high in Butula and less intense in Busire and Rachar. This could be attributed to build-up of inoculum in soil and plant debris due to continuous bean cropping and poor field sanitation (Mwangombe *et al.,* 2007; Gichangi *et al.*, 2012). Further, Akhavan *et al.* (2013) and Tuti *et al.* (2015) also reported the influence of geographical area, climate, cultural practices, host genotypes and bacterial strains on the survival of the pathogens in soil or plant debris and severity of angular leaf spot. In addition, Sengooba and Mukiibi (1986) investigating the role of seed inoculum and debris infestation in disease development by *Xanthomonas axanopodis* p.v *phaseoli,* reported dry leaf debris as sources of viable inocula for several years.

Foliar diseases were more severe on bean crop raised from farm saved and market sourced KATX56, KATX69 and GLP2 compared to KK8 and certified GLP2 crops. Most foliar diseases are seed borne suggesting that farm saved and market sourced GLP2 and KATX69 seeds were of poor quality and had high seed-borne inoculum levels as shown in this study hence the high disease transmission. These findings agree with Sabry *et al.* (2013) and Coomes *et al.* (2015) who reported high transmission of bacterial and fungal diseases in farm saved or unclean certified seeds. Moreover, Sharma *et al*. 2009 reported 75% yield loss in crop raised from infected seed compared to 57% loss from other inoculum sources. Despite being raised from certified seed, both KK8 and GLP2 crops had some disease infections. This agrees with reports by Karavina *et al.,* (2008) who stated that certified seeds compared to farm saved seed, had less inocula of *Xanthomonas axonopodis* p.v *phaseoli*. Oshone *et al.* (2014) isolated bacterial and fungal pathogens from bean seed samples obtained from small holder farmers in Eastern Ethiopia and revealed certified seed samples to have contamination with bacterial pathogens although less compared to farm saved seed. Most fungal and bacterial pathogens are seed borne and seed is the main source of inocula (Allen *et al*. 1998, Islam *et al.,* 2009). This suggests that seed certification standards as in case of certified KK8 and certified GLP2 reduces inoculum levels leading to low disease pressure compared to uncertified seed. Varietal susceptibility has also been reported as a reason for increased disease intensity in susceptible cultivars (Icishahayo *et al.,* 2009).

4.5.3 Yield and yield components

The potential yield per hectare in Kenya ranges between 1400kg/Ha to 2000Kg/Ha (FAOSTAT (2015). The results of this study showed that none of the varieties achieved the potential bean yield. Beans planted in Arongo had the highest yield performance unlike bean yields in Butula

which had the lowest. Low yields could be attributed to several factors including poor soil fertility and high prevalence of fungal and bacterial. All the soils in the study sites were deficient in nitrogen, soil carbon and some in phosphorus. In addition, prevalence of fungal and bacterial diseases in Butula led to reduced yields. These findings agree with Otsyula *et al.* (1998), Lunze *et al.* (2012) and Mulei and Woomer (2015) who reported low bean yields due to poor soil fertility, high disease pressure, insect pest infestations and poor agronomic practices.

Both variety KATX56 and certified variety GLP2 had high seed and biomass yields and number of pods unlike farm saved variety GLP2 which yielded poorly. This could be explained by high disease pressure in farm saved variety GLP2 crop due to poor quality and high bacterial contamination of seed used to raise the crop consequently causing yield losses. The good performance of variety KATX56 could be due to its quality attributes such as drought tolerance and high yielding (Karanja *et al.,* 2010, Gichangi *et al.,* 2012). The results concur with Sibiko (2012) who reported positive influence of good quality and certified seed on bean productivity. Oshone *et al.,* (2014) further reported that most common fungal diseases are associated with seeds retained by farmers (own saved seeds) and those sampled from markets and cooperative unions compared to certified seed leading to decline in productivity in bean crops raised from such seed sources. However contrary observations were made by Filho *et al.* (1997) who reported that severity of angular leaf spot disease did not relate with yield reduction.

High proportions of discoloured, shrivelled seed, infected seedlings and high bacterial pathogen inoculum in seed resulted in increased intensity of foliar diseases on the bean crops which consequently reduced bean yield. This agrees with Allen *et al*. (1998) who reported that seed is the primary source of inocula for bacterial pathogens causing high severity of foliar diseases. Additionally, foliar diseases causes huge crop losses, reduction in plant growth and productivity

of crops (Dawson and Bateman, 2001; Islam *et al.,* 2009). The findings in this study indicates that low soil nutrients levels of N, P, K and C lead to decreased bean yields. The findings further indicated that high intensity of fungal and bacterial diseases on the bean crops in the field lead to reduced bean yield. This concurs with the findings by Zingore and Giller (2012), who reported strong positive correlation between yields of soybean and soil organic matter. The findings further corroborate with findings by El-Bramawy and Shaban (2010) who reported decreased disease incidences in faba beans with increase in soil fertility. Other studies by Leite, *et al.* (2006) and Lemessa, *et al.* (2011) reported negative effect of foliar diseases such as ALS, WB, CBB on bean and sunflower yield. High Bacterial contamination in seed leads to high seedling infections and increased intensity of foliar diseases on beans which consequently reduced bean yields. Production is a function of photosynthesis in the leaves and therefore, foliar diseases cause defoliation that leads to reduction of the leaf area required for photosynthesis hence low yields (Jesus Junior *et al.,* 2003). Low bean yields are attributed to high disease pressure low soil fertility levels in the respective sites.

CHAPTER FIVE: CONCLUSION AND RECOMMENDATION

5.1 Conclusion

Sites with poor soil fertility had high populations of soil borne pathogens and consequently high incidence of root rots infection and disease intensities. This indicates that Low soil organic matter and poor soil properties as in LM4 play a significant role in increasing the severity of soil borne pathogens. Poor soil fertility and build-up of pathogens are as a result of nutrient depletion due to allocation of small pieces of land to continuous bean production without fertilization and lack of crop rotation by most farmers. Favourable weather conditions, poor drainage and soil compaction as in LM4 also contributed to further increase in the severity of root rots and yield losses**.**

Different levels of root rot infection and intensities were recorded in all the seed varieties. Root rot infection and intensities were high on farm saved and market sourced seeds while moderate for certified seeds but lower for certified seeds of tolerant varieties. This suggests that seed plays a role in disseminating root rot causing pathogens as shown by the high disease infection and intensity levels on crops raised from farm saved and market seed as a result of build-up of inoculum in recycled seed. Yields were also higher in crops raised from certified seed of tolerant varieties which showed low effects of root rot. This shows that use of tolerant varieties considerably reduce effect of root rot hence improved bean yields. Use of clean seed and tolerant varieties to both biotic and abiotic stresses among farmers should be promoted to improved bean production.

Farm saved and market sourced seed had low purity and germination rate resulting to poor seed health. An indication that farm saved and markets seed are of poor quality and are source of primary inoculum for subsequent seasons. High population of seed borne bacterial pathogens were isolated from farm saved and market sourced seeds before planting and at harvest regardless of the

variety. This suggests that there was already available bacterial inoculum in farm and market seed due to continuous recycling and build-up of inoculum in seed.

There was high prevalence of both fungal and bacterial diseases of common bean in Butula and lower in Busire. This implies that improved soil fertility and good agronomic practices like field sanitation and rotation by farmers play a role in reducing disease pressure in the respective site. Crops raised from certified GLP2 and KK8 seeds had low disease intensity unlike crops raised from farm saved, market sourced GLP2 and KATX varieties implying that most fungal and bacterial disease are seed borne and that inoculum in seed is responsible for increased disease pressure. It also means that certified seed is disease free and genetically pure therefore low disease transmission and infection. Certified seed production is done with genetically pure seed, in diseases free soils where routine field inspections, proper handling at harvest and seed certification processes are done before releasing to stockist. Farm saved seed on the other hand, is produced using impure and already infected seed or using a clean seed but in already contaminated soil or in fields with infected debri. Improper handling of seed during harvesting process and storage may also cause additional damages to the seed resulting to poor quality of such seed. The findings in this study suggest the need to make available to farmers cost effective certified seed which are disease free and tolerant to the harsh climatic conditions present in most parts of the country especially Western Kenya.

5.2 Recommendations

i. Farmer training on the use of certified clean seed and seed management through sorting to remove discoloured / shrivelled seeds, insect damaged seeds in order to reduce disease inoculum for subsequent season.

ii. Adoption of soil fertility and disease management measures such as crop rotation, seed treatments with fungicides and field sanitation and liming of acidic soil.

iii. Further research is recommended to establish the relationship between soil fertility levels and soil borne pathogen inoculum densities.

iv. Subsidization and price regulation of commercial certified bean seed production to ensure availability of affordable certified clean bean seed to small scale resource-poor farmers.

v. Farmer training on production, processing, storage and use of their own clean seed to minimise seed transmission of diseases.

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APPENDICES

Appendix I: Average temperature (°C), precipitation (mm) and relative humidity (%) in Kakamega in 2015

Month	Total precipitation	Maximum temperature	Relative humidity 06Z	Relative humidity 12Z
January	3.4	31.0	60.0	31.0
February	52.2	32.6	60.6	31.2
March	210.6	32.4	55.2	33.0
April	368.3	28.0	83.2	65.4
May	302.5	27.6	84.0	66.0
June	230.7	26.9	87.9	64.4
July	146.0	28.1	82.0	54.0
August	198.8	28.8	77.0	52.0
September	110.5	28.8	77.0	57.0
October	195.6	28.2	*	*
November	*	*	*	*
December	132	27.6	*	*

Appendix II: Average temperature (°C), precipitation (mm) and relative humidity (%) in Kisumu in 2015

Month	Total precipitation	Maximum temperature	Relative humidity 06Z	Relative humidity 12Z
January	3.5	30.7	57.0	40.0
February	53.4	33.0	53.0	35.0
March	205.9	33.6	54.0	34.0
April	284.9	28.6	81.0	58.0
May	175.3	28.6	78.0	59.0
June	49.8	28.3	76.0	55.0
July	84.9	29.9	69.0	47.0
August	21.7	30.9	62.0	43.0
September	154.5	30.2	64.0	48.0
October	200.3	30.5	*	*
November	143.6	28.6	*	*
December	218.9	28.1	*	*