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Effect of incorporating lablab biomass in soils on root rot disease complex and yield of beans intercropped with maize

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ABSTRACT
Root rot is a major constraint to bean production in western Kenya causing poor crop stand and high yield losses. The disease is caused by a complex different pathogens which together exhibit synergistic effects. The pathogens survive in soils as resting spores and options of managing the disease are limited This study evaluated the effectiveness of incorporating lablab biomass in managing root rot of beans in maize intercrop system. The experimental treatments evaluated were incorporation of lablab biomass over the whole plot, biomass placed between rows of beans, biomass removed from the plot plus application of inorganic fertilizer and biomass removed from the plot without fertilizer application. In each experimental treatment, four bean varieties KK8, KK15, KK072 (tolerant to root rot) and GLP2 (susceptible to root rot) were planted intercropped with maize. The experiment was carried out at two agro-ecologically and soil fertility diverse sites. Incidence of root rot and chafer grub was determined at early growth stages while biomass and yield of both maize and beans were determined at harvest. Incorporation of lablab biomass increased soil carbon and nitrogen content but reduced both pH and cation exchange capacity. However, it reduced root rot incidence for the root tolerant bean varieties at the low soil fertility site but increased chafer grub incidence. Bean stem bases from the high soil fertility site had higher incidences of infection with *Fusarium oxysporum* while *Fusarium solani* and *Macrophomina phaseolina* were more prevalent in the low soil fertility site. Addition of lablab biomass significantly increased both bean biomass and seed yields in both low and high soil fertility sites. The positive effect on yield was more pronounced at the low soil fertility site. The study indicated that addition of lablab biomass to soils is beneficial in managing root rot of beans and improving crop yields in low soil fertility areas. The use of green manure soil amendments is an ecologically sustainable way of increasing bean yield for small scale farmers.

***Keywords*:** *Chafer grub, Phaseolus vulgaris, root rot complex, soil fertility, yield*

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is an important source of dietary protein and component of production systems among resource-poor farmers in the developing countries (Katungi *et al*., 2010; Mwang'ombe et al., 2008). However, root rot caused by *Fusarium* species is a major constraint to bean production in the tropics and in western Kenya the disease has been reported to cause total crop failures (Nzungize et al., 2012; Otsyula et al., 2003). Root rots are caused by a complex of soil-borne fungal pathogens which include *Fusarium solani* f.sp. *phaseoli*, *Pythium ultimum* var. *ultimum*, *Macrophomina phaseolina*, and *Rhizoctonia* spp (Mwang'ombe et al., 2008; Nzungize et al., 2012). Co-occurrence of the different pathogens exhibit synergistic interactions resulting in higher disease incidences and severity (Ongom et al., 2012). These fungi survive in soil organic matter as saprophytes or as resting spores (Waller and Brayford, 1990). The long-term persistence of the survival structures of the fungi makes it difficult to manage the disease complex. The pathogens mainly attack plants that have been weakened by chafer grubs (*Cyclocephala* spp.), bean fly (*Ophiomyia phaseoli*) infestation, nematodes and other pests (Medvecky *et al*., 2007). There are limited options for managing root rot disease complex of beans mainly because chemical fumigation is not feasible and complete genetic resistance is non-existent. Only a few bean varieties are tolerant and they only reduce the effects of the disease through development of adventitious roots above the point of infection on the stem bases (Abawi et al., 1985). Other limitations to conventional

methods of managing root rot pathogens include harmful effects on environment and human health, development of resistance by plant pathogens and lack of varieties that are tolerant or resistant to the multiple disease causing pathogens (Ha and Huang. 2007; Nzungize et al., 2012).

Agronomic practices such as crop rotation, deep tillage, planting of fallow crops and application of organic amendments have been shown to have positive changes in soil structure and root rot disease dynamics leading to increase in yields (Bailey and Lazarovits, 2003; Tan and Tu, 1995). These practices reduce disease inoculum density in the soil, deprive the pathogen of its host and create conditions that favour the growth and development of microorganisms that are antagonistic to plant pathogen (Peter *et al*. 2009; Meenu *et al*. 2010). Addition of soil organic amendments increases the population of antagonistic microorganism such as *Trichoderma harzianum, Bacillus* spp., *Pseudomonas fluorescens* (Ha and Huang, 2007). The addition of organic matter and crop biomass influence disease pathogen viability and distribution through the release of allelochemicals (Bailey and Lazarovits, 2003).

Leguminous crop biomass and green manures improve soil fertility, increase nutrient supply in the soil through biological nitrogen fixation and improved soil structure (Toomsan *et al*., 2000). They however increase the abundance of root rot feeding chafer grubs (Medvecky *et al*., 2006). Chafer grubs are abundant in soils with high organic matter and they feed on roots just below the soil level (Abile and Alemayehu, 2014). The grubs affect all field crops and heavy infestations result in poor crop stand and may reduce yields by up to 20%. *Lablab purpureus* is a leguminous crop rich in nitrogen and useful as an organic amendment because it produces large amounts of biomass, has rapid establishment and its soft stem makes it easy to chop prior to incorporation (Mureithi *et al*., 2003). Farmers in Western Kenya have been introduced to new green manure legumes for soil fertility enhancement but little is known about the effect they have on soil-borne pests and diseases (Medvecky *et al*., 2007). This study therefore aimed to determine the effect of lablab biomass on soil borne bean diseases.

MATERIALS AND METHODS

Experimental site

Field experiments were carried out in farmers' fields in Koibem and Kapsengere in Nandi South sub-County. Koibem is high soil fertility while Kapsengere is a low soil fertility area, based on the time since the land was opened for cultivation from Kakamega forest (Nyberg *et al*., 2012). Koibem has been under cultivation for 5-30 years while Kapsengere has been under cultivation for 80-105 years (Odundo *et al*., 2010). Nandi South sub-County lies within latitudes 0° and 0°34'' North and longitudes 34°44'' and 35°25'' East at an elevation of 1850-2040 m above sea level (Nyberg *et al.,* 2012). Annual precipitation is 1200 mm to 2000 mm with temperature ranging from 18° to 25°C. Soils are mainly well drained clay-loams, classified as Nitosols (FAO-UNESCO, 1988). The sub-County's main agro ecological zones are upper highlands (UH) covering about 5%, lower highlands (LH) covering 24% and upper midlands (UM) covering 56% of the total land area (Jaetzold, 2006).

Experimental design and layout

Lablab variety Rongai was planted during the short rains in 2011 at both Koibem (high soil fertility site) and Kapsengere (low soil fertility site). The spacing was at 45 cm x 30 cm and the crop was allowed to grow until flowering when the vegetation was harvested and biomass used for incorporation into the soil during the subsequent long rain season in 2012.. The vegetative mass of the lablab crop was harvested, the vines chopped into small pieces for ease of handling during incorporation and oven dried to constant weight. The biomass experimental treatments were: lablab incorporated over the whole plot, lablab biomass placed between rows of beans, lablab biomass cut and removed from the plot, lablab biomass cut and removed and di-ammonium phosphate (DAP) fertilizer applied at 130kg/ha. The chopped lablab biomass was applied to the appropriate plots measuring 5 m x 4 m at the rate of 50 kg/ha. Four bean varieties: KK8 (tolerant to root rot), KK15 (tolerant to root rot), KK072 (tolerant to root rot and bean fly) and GLP2 (susceptible to root rot and bean fly) were used for each lablab biomass treatment. Each bean variety was planted on 5 m x 4 m plots intercropped on the same row with maize at spacing of 75 cm x 15 cm, between and within rows respectively. Maize was planted at a spacing of 75 cm by 30 cm. The plots were separated by 1m paths and the treatments laid out in a randomized complete block design with split plot arrangement. Bean varieties comprised the main plots and lablab biomass management treatments comprised the subplots. Each of the lablab biomass treatments/ bean variety combination was replicated in three blocks. Data collected included soil nutrient content before and after incorporation, incidence of root rot, incidence of chafer grub damage, plant biomass and seed yield.

Assessment of root rot incidence and infection of bean stem bases

Incidence of root rot infected bean plants was determined by counting the number of plants

showing root rot symptoms per plot at the second, fourth and sixth week after emergence. Root rot infected plants were recognized based on symptoms such as yellowing of leaves, wilting, stunted growth, death and brown discoloration on the roots. At the fourth week after emergence, five plants showing root rot symptoms were sampled from each plot. Each stem base was washed under running tap water and cut into five one centimeter pieces that were surface sterilized in 2.5% sodium hypochlorite and rinsed in sterile distilled water. Five stem base pieces were aseptically plated in each Petri dish containing potato dextrose agar (PDA) amended with 50 ppm streptomycin sulphate antibiotic to suppress growth of bacteria. After 7 to 14 days the number of stem base pieces showing fungal growth in each plate was counted and each fungal colony type was identified based on colony colour, type of growth, colour of underside of the colony and spore morphology (Nelson *et al*. 1983).

Assessment of chafer grub damage incidence

White chafer grub (*Cyclocephala* spp.) damage incidence was determined by counting the number of plants showing grub damage and the presence of C-shaped larvae of scarab beetles at seedling stage. The suspected grub damaged plants were dug up and roots examined for signs of pruning and removing the larvae from the root one of the damaged seedling. The number of damaged plants and the number of grubs was counted.

Assessment of yield and yield components

Yield parameters assessed were biomass at harvest, seed yield and maize cob weight per plot and the data was extrapolated to kg/ha. At pod maturity, the bean crop was harvested, shelled and biomass and seed weight determined for each plot. Yield for the maize crop was determined at hard dough stage by taking the weight of cobs and stover biomass from each plot.

Statistical data analysis

All data was subjected to analysis of variance (ANOVA) using Genstat software version 7.1 (Payne et al., 2008) and means were separated using Student-Newman-Keuls (SNK) test at P=0.05.

RESULTS

Effect adding lablab biomass on Soil nutrient status

Addition of lablab biomass significantly increased the percent organic carbon and nitrogen in both experimental sites (Fig 1).

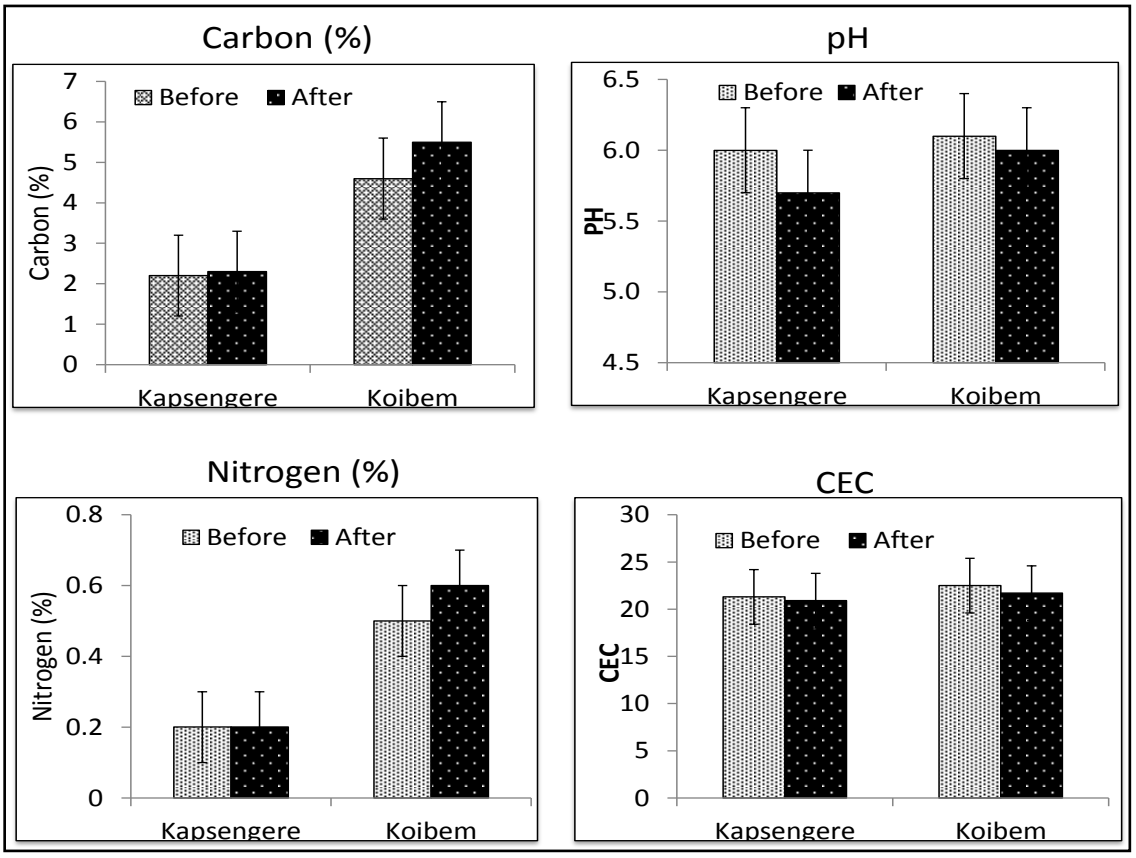

Fig.1. Soil organic carbon, nitrogen, pH and CEC before and after addition of lablab biomass at two sites in Nandi County during the 2012 long rains season

The increase in carbon and nitrogen was greater in Koibem (high fertility site) compared to Kapsengere (low fertility site). The soils in Koibem site already had significantly higher carbon and nitrogen before amendment with lablab biomass. However, the addition of the lablab biomass reduced both pH and cation exchange capacity (CEC) in both sites. The sites did not significantly differ in both pH and CEC.

Effect of lablab biomass on root rot incidence and stem base infection

The mean root rot incidence was insignificantly different in the two sites (p=0.8787). However, there was an interaction effect between the variety and site and also between management method and the site. Removal of lablab biomass led to significantly higher incidences of root rot in the low soil fertility Kapsengere site while in the high soil fertility Koibem site there were no significant differences among the treatments. Bean variety GLP2 had the highest root rot incidence at the low soil fertility Kapsengere site, but in the high soil fertility Koibem site there were no significant differences among the bean varieties (Fig 2).

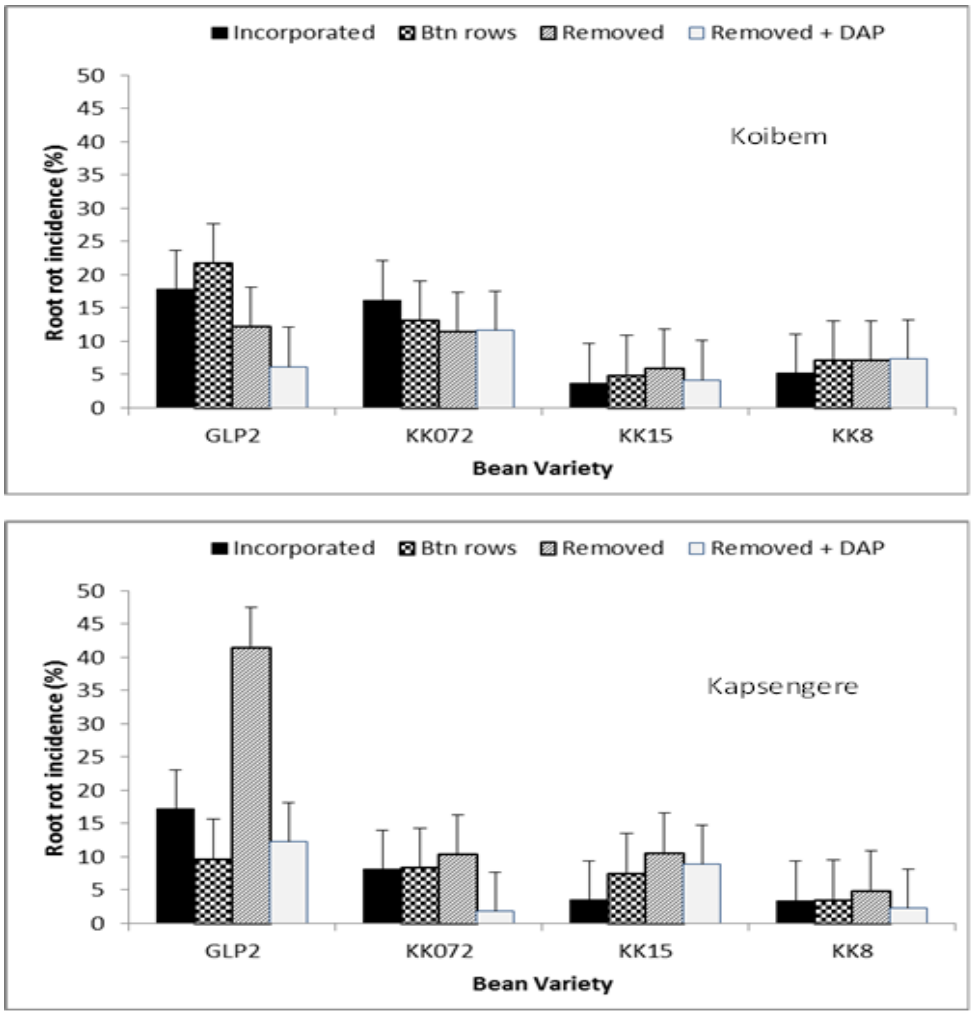

Fig. 2: Percentage root rot incidence on four bean varieties under different lablab biomass management methods in Kapsengere (low soil fertility) and Koibem (high soil fertility) in Nandi South.

The tolerant bean varieties KK072, KK15 and KK8 generally had low incidences of root rot compared to the susceptible GLP2 variety. Incorporation of lablab biomass resulted in lower root rot incidences for the root rot tolerant bean varieties KK15 and KK8 compared to plots where lablab biomass had been removed.

The main root rot pathogens isolated from the bean stem bases were *Fusarium oxysporum*, *Fusarium solani* and *Macrophomina phaseolina* (Table 1). Most bean stem bases from the high soil fertility Koibem site had higher incidences of infection with *Fusarium oxysporum* compared to those from the low soil fertility Kapsengere site. However, *Fusarium solani* and *Macrophomina phaseolina* were more prevalent in the low soil fertility Kapsengere site. Incorporation of lablab biomass did not have a significant effect on the percentage number of bean stem bases infected with the different root rot pathogens. Incorporation of lablab biomass over the whole plot significantly reduced the incidence of bean stem base infection in the low soil fertility Kapsengere site.

Table 1. Percentage number of bean stem bases infected with different root rot pathogens under different biomass management methods in two sites in Nandi South

	Kapsengere (low fertility)			Koibem (high fertility)		
	Foxy	Fsol	Mpha	Foxy	Fsol	Mpha
Incorporated	73.9_a	10.6_b	29.4_a	86.7_a	5.6_a	1.7_a
Between rows	55.6_c	33.9_a	17.8_b	80.0_{ab}	8.3_a	1.7_a
Removed	64.6_b	31.1_a	36.1_a	82.8_a	10.6_a	2.8_a
Removed + DAP	52.8_c	35.0_a	18.9_b	89.4_a	5.0_a	6.7_a
LSD $_{(p\leq0.05)}$	8.6	8.7	6.8	8.6	8.7	6.8
C.V (%)	27.7	104.8	116.5	27.7	104.8	116.5

Foxy = *Fusarium oxysporum*, Fsol = *Fusarium solani*, Mpha = *Macrophomina phaseolina*; LSD = least significant difference; C.V = coefficient of variation; DAP = Diammonium phosphate fertilizer

Effect of lablab biomass on chafer grub incidence

There was a significant interaction effect between variety and biomass management method (P=0.005; Table 2). Treatments where lablab biomass were uniformly incorporated over the whole plot had the highest chafer grub incidence in the low soil fertility Kapsengere site, while low grub incidence was observed in plots where the biomass was removed and where it was removed and fertilizer applied. There were no significant differences among the treatments in the high soil fertility Koibem site (Table 2). There were higher incidences of chafer grub in the low soil fertility Kapsengere than in the high soil fertility Koibem site. Bean variety GLP2 had the highest incidence of chafer grubs in all treatments and varieties KK15 and KK8 had the lowest.

Table 2: Percentage Chafer grub incidence under different biomass management methods in two sites in Nandi south

Site/Variety	Incorporated	Btn rows	Removed	Removed+DAP	Mean
Kapsengere (low soil fertility site)					
GLP2	3.7_a	2.8_b	1.6_c	1.9_c	2.5
KK072	2.3_a	1.6_b	1.2_c	1.9_b	1.8
KK15	2.6_a	0.4_b	0.0_c	0.0_c	0.8
KK8	0.8_b	1.7_a	0.4_c	0.0_d	0.7
Mean	2.3_a	1.6_b	0.8_c	1.0_c	1.4
Koibem (high soil fertility site)					
GLP2	2.8_b	3.0_a	1.3_d	2.1_c	2.3
KK072	1.4_a	1.4_a	0.5_b	0.6_b	1.0
KK15	0.0_a	0.0_a	0.0_a	0.0_a	0.0
KK8	0.4_b	1.6_a	0.0_c	0.4_b	0.6
Mean	1.1_b	1.5_a	0.4_d	0.8_c	1.0
LSD site =0.2, LSD Mgt= 0.3, LSD variety =0.5, LSD V x Mgt =0.7, CV (%) =49.5					

LSD: Least significant difference at 5% level, CV: Coefficient of variation, Btn: between, DAP: Diammonium phosphate, Mgt: Management,Var: Variety

Effect of lablab biomass on bean seed yield and biomass

The interaction of variety and lablab biomass management method was significant (P=0.002) for bean seed yield (Table 3). In addition, significant differences were observed among the varieties and lablab biomass management methods (Table 4).

Table 3: Root rot and chafer grub incidence, bean seed yield and biomass under different lablab biomass management methods

	Lablab biomass management method			
Site/Variety	Incorporated	Btn rows	Removed	Removed+DAP
Kapsengere (low soil fertility site)				
Root rot	8.0_{b}	7.0_{b}	17.0_{a}	6.0_{b}
Chafer grub	2.0_{a}	2.0_{a}	1.0_{b}	1.0_{b}
Biomass	157.1_{b}	126.5_{bc}	108.8_{c}	267.0_{a}
Seed yield	245.0_{b}	213.9_{b}	155.8_{c}	318.7_{a}
Koibem (high soil fertility site)				
Root rot	11_{a}	12.0_{a}	9.0_{a}	7.0_{a}
Chafer grub	1.0_{b}	2.0_{a}	0.0_{c}	1.0_{b}
Biomass	161.1_{bc}	176.8_{b}	145.0_{c}	239.0_{a}
Seed yield	153.8_{ab}	142.0_{b}	120.8_{b}	192.6_{a}

Values followed by different letter(s) along samerow are significantly different ($P \leq 0.05$)

Table 4: Root rot and chafer grub incidence, bean seed yield and biomass for different bean varieties

	Bean variety			
Site/Variety	GLP2	KK072	KK15	KK8
Kapsengere (low soil fertility site)				
Root rot	20.0_{a}	7.0_{b}	8.0_{b}	4.0_{b}
Chafer grub	3.0_{a}	2.0_{b}	1.0_{c}	1.0_{c}
Biomass	33.0_{c}	172.6_{b}	213.8_{a}	239.9_{a}
Seed yield	51.8_{c}	216.8_{b}	351.3_{a}	313.4_{a}
Koibem (high soil fertility site)				
Root rot	15.0_{a}	13.0_{a}	5.0_{b}	7.0_{b}
Chafer grub	2.0_{a}	1.0_{b}	0.0_{c}	1.0_{b}
Biomass	82.4_{c}	110.3_{c}	329.2_{a}	200.0_{b}
Seed yield	41.9_{c}	53.8_{c}	408.3_{a}	105.1_{b}

Values followed by different letter(s) along the same row are significantly different at 5% level of probability

Removal of lablab biomass and application of fertilizer led to the highest bean yield in both the low (Kapsengere) and high (Koibem) soil fertility sites (Table 3 and 4). However, the lowest seed yield was attained at both sites in plots where lablab biomass was removed and no fertilizer applied. Addition of lablab biomass significantly increased both bean biomass and seed yields in both low (Kapsengere) and high (Koibem) soil fertility sites. Bean variety GLP2 had the lowest seed yield in both Kapsengere and Koibem while varieties KK8 and KK15 had the highest yield. Significantly higher seed yields were attained in the low fertility (Kapsengere) site compared to the high (Koibem) fertility site. The interaction between variety and biomass management method was highly significant (P<0.001) for biomass at harvest. In addition, significant differences were observed among the varieties and biomass management methods (Tables 3 and 4). Root rot susceptible bean variety GLP2 had the lowest biomass in Kapsengere, while there were no significant differences among the other three bean varieties. The highest crop biomass in both sites was attained where biomass was removed and fertilizer applied, while the lowest crop biomass was in plots where lablab had been removed.

Effect of lablab biomass on maize yield

Lablab biomass management method was highly significant (P<0.001) for maize stover and cob weight (Fig 3). The highest cob weight in both sites was attained in plots where DAP fertilizer was applied, followed by plots where lablab biomass were incorporated over the whole plot and where they were placed between rows of bean. Removal of lablab biomass led to the least cob weight in both sites (Fig 3). Lablab biomass management method was highly significant (P<0.001) for stover biomass. Addition of fertilizer led to the highest stover

biomass in both sites, while removal of lablab biomass led to the lowest stover biomass. There was no significant difference between treatments where lablab biomass was incorporated uniformly over the whole plot and where DAP fertilizer was applied in Kapsengere (Figure 3). The high soil fertility Koibem site had significantly higher cob weight and stover biomass than the low soil fertility Kapsengere site.

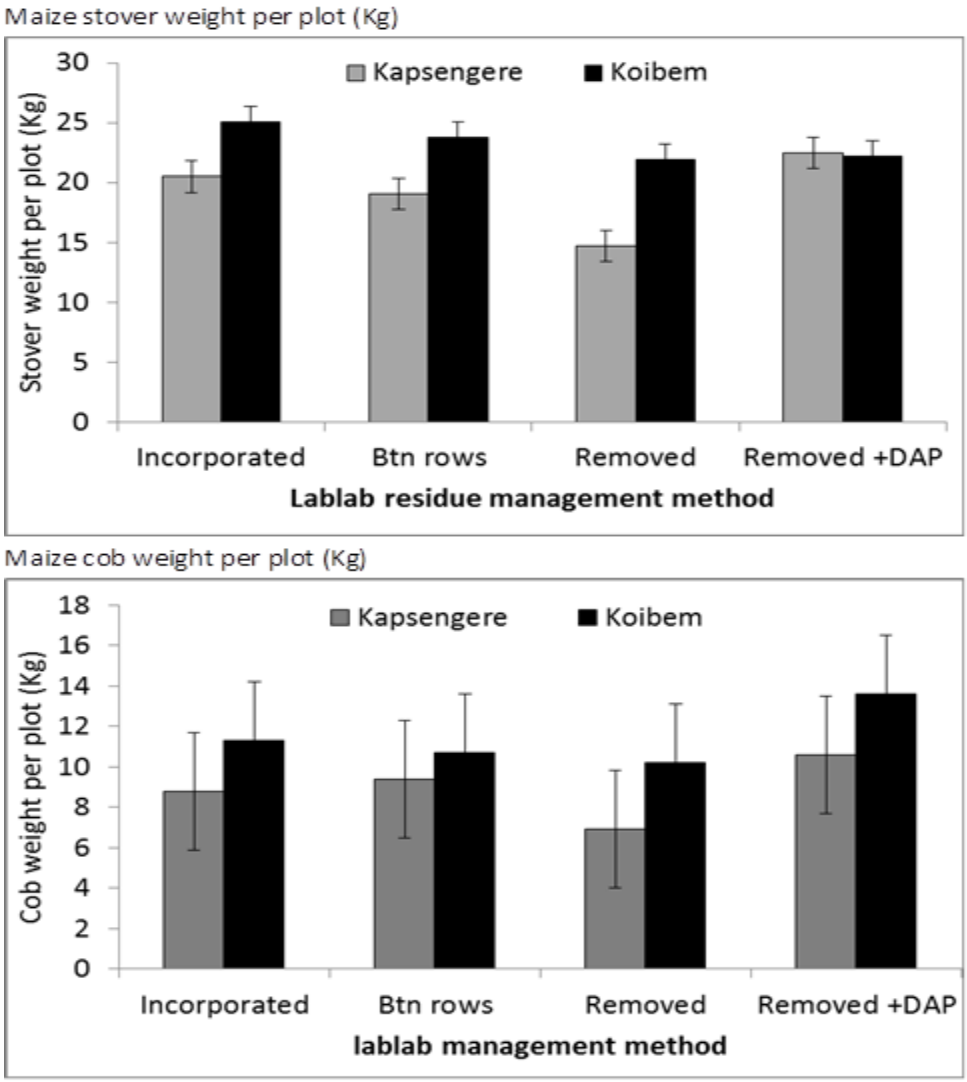

Fig.3. Maize stover and cob weight per plot (Kgs) under different lablab biomass management methods in Kapsengere (low fertility) and Koibem (high fertility) sites in Nandi south

DISCUSSION

The results of the study showed that addition of lablab biomass to soils before planting beans reduces root rot incidence in low soil fertility sites especially for the root rot tolerant bean varieties. The biomass, however, increased chafer grub infestation. Removal of lablab biomass led to the highest root rot incidence The results are in agreement with other previous studies by Medvecky *et al*. (2007), Abawi and Widmer (2000), Peters *et al*. (2003) and Okoth and Siameto (2010). However, Medvecky *et al*., (2007) reported that retention of lablab biomass increased *Pythium* seed infection. The lower levels of root rot in plots with DAP fertilizer application is because inorganic fertilizer improves the vigour of the crop and therefore enables it to overcome the effects of the root rot pathogens (Duffy and Defago, 1999). Increase levels of phosphates have been shown to reduce root rots (Bailey and Lazarovits, 2003).

Suppression of disease causing plant pathogens by addition of crop biomass can be explained by the production of allelochemicals during microbial decomposition (Bailey and Lazarovits, 2003). Addition of crop biomass changes microbial the balance of beneficial and pathogenic microorganisms in the soil resulting in improved productivity (Mathre et al., 1999). The mechanisms involved in suppression of disease causing pathogens include reduced ability of the pathogen to survive, antibiotic production, parasitism, increase in competition from antagonistic microorganisms, reduced spore germination due to increased soil carbon content and systemic acquired resistance (Bailey and Lazarovits, 2003; Bareja et al., 2010; Stone et al., 2003). Organic amendments containing high amounts of nitrogen have greater disease pathogen suppressive effects due to ammonia liberated during decomposition (Bailey and Lazarovits, 2003; Tenuta, 2001). The pathogen suppressive effects are greater in soils that accumulate ammonia and the accumulation of the disease-suppressive benefits is slow and increases over years (Tenuta, 2001).

The plots where lablab biomass uniformly incorporated over the whole plot had the highest chafer grub incidence in both sites and those where biomass were removed the lowest. This agrees with Medvecky *et al*., (2006); (2007) who found out that lablab biomass increased chafer grub incidence due to increased soil fertility and favourable conditions for oviposition and grub survival. Chafer grubs are favoured by high organic matter, especially in loose soils with moderate to low rainfall (References).

The results of this study show that incorporation of lablab biomass improved biomass and seed yields of both bean and intercrop maize. The results agree with results by Belachew and Abera (2011) and Shah *et al*., (2011) who reported that green manure significantly increased wheat yields. Mureithi *et al*., 2003 and Tolanur (2009) also reported that addition of organic together together with inorganic fertilizers increased grain and straw yield of chick pea without deterioration of soil quality. The improved yields on plots where lablab biomass was added can be explained by the corresponding increase in nitrogen and soil organic matter. Lablab biomass has a nutrient composition of 3.2% N, 0.21% P, 1.57% K and 0.2% Mg (Lelei, 2004; Nworgu and Ajayi, 2005) and therefore the increase in soil nutrient status. Addition of lablab biomass also increase root growth due to increased release of nutrients by cellulolytic and hemicellulolytic actinomycetes that degrade the organic matter (Bareja et al., 2010; Nitta, 1991). The improved soil nutrient status also improves nodulation and plant growth thus contributing to better yields as a result of increased nitrogen fixation (Bareja et al., 2010). These results conform to findings of studies done by Ayuke *et al*, (2004), Njeru *et al*., (2007) and Odhiambo (2011) which showed that green manure and plant biomass increase maize yield comparably to inorganic fertilizer.

The study indicated that addition of lablab biomass to soils is beneficial in managing root rot of beans and improving crop yields in low soil fertility areas. The associated negative effect of increase chafer grub incidences is however countered by the improved crop vigour and use of root rot tolerant bean varieties which had reduced level of damage. This integrated soil nutrient and disease management method is an ecologically sustainable way of increasing bean yield for small scale farmers.

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