

## Effect of Lablab Green Manure on Population of Soil Microorganisms and Establishment of Common Bean (*Phaseolus vulgaris* L.)

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**Abstract:** Green manures improve soil health and fertility but application of undecomposed lablab residues lead to low crop establishment. The study was carried out to determine the effect of green manure on microbial population and establishment of bean crop. Bean varieties were planted on plots each treated with lablab green manure at one ton ha<sup>-1</sup> over whole plots and in rows, DAP fertilizer was applied at 75 kg/ha. Data was collected on microbial population, crop emergence, root rot incidence and severity, and yield. Green manure incorporation increased soil organic carbon, nitrogen, phosphorus and potassium but reduced germination percentage by about 35% and increased incidences of root rot by 30% compared to plots without green manure. The population of root rot pathogens was significantly higher in plots treated with lablab green manure reduced grain and biomass yields by 25%. Green manure increases soil nutrients directly and improves crop establishment after decomposition. Results of the study revealed a considerable increase in the population of root rot pathogens with corresponding decrease in the population of antagonistic fungi thus the poor emergence and crop establishment can be associated with increase in population of root rot pathogens and stress experienced by seeds during decomposition

Keywords: Green Manure, Phaseolus vulgaris, Root Rot, Soil Health

## 1. Introduction

Common bean is an important source of dietary protein and calories, iron and supplementary amino acids [1]. It is widely grown in Kenya and is only second to maize and is grown for its green leaves, green pods, and immature or dry seeds. In Kenya, beans are largely grown by small scale farmers with fewer than five acres and mostly intercropped with maize [2]. The crop, however, is grown under challenging conditions, including marginal lands with infertile soils prone to drought, pests, and diseases [3]. Drought and low soil fertility are primary constraints to crop production in most of the third world countries [4]. The decline in soil fertility has prompted farmers to embrace the use of inorganic fertilizers to rejuvenate the soil, however, they are expensive [5]. There is need for farmers to use alternative means to restore soil productivity by the use of green manure as a replacement for nitrogen required fertilizers [6].

Green manures contain lignin, cellulose, hemicellulose, micro and macro-nutrients and their decomposition is dependent on lignin, cellulose content and carbon: nitrogen ratio, which is reliant on the type of the crop, the environment and the conditions of [7]. Residues containing high contents of lignin decompose slowly than those with low lignin content [8]. When green manure residues decompose, they are converted into carbon dioxide, water, nitrogen, sulphur oxide and therefore naturally replenishes the carbon, nitrogen and sulphur cycles [9]. The process of decomposition is mediated by microbial activities [10] where bacteria, fungi, molds, protozoa, actinomycetes pray on the decaying organic materials in initial stages and millipedes and centipedes and earthworms act on the materials on the later stages [11]. During breakdown, energy is released in the form of heat in oxidation of carbon to carbon dioxide, and water. Microorganisms decompose the organic materials for energy used for growth and carbon for the synthesis of new cell material [12]. The products that are released during decomposition may be toxic to plant and enhance pathogenic fungi affecting crop germination and establishment resulting in reduced density and vigour [13], however, the toxic effects of these products depend on their concentrations in the soil.

Green manure toxicity is common in the early stages of crop development, resulting into reduced germination, root growth and seedling weight and height [14]. Inhibition of germination may be due to release of leachates, reduced nitrate because of nitrogen tie up by the soil microbes and promotion of seed and seedling pathogens [15]. The poor emergence and establishment may be due to provision of food source for many pathogens by the fresh residues incorporated in the soil [16] thereby increasing infections caused by root rot fungal pathogens. Many experiments have indicated increased level of disease may be a contributing factor to the effect on crop diseases. If susceptible bean seeds are planted they are invaded by the high population of pathogenic fungi thereby causing poor germination and establishment of common beans. Proper characterization of the composting products is necessary to predict the harmful effects of green manure applications to the and therefore to reduce harm to crops. The study was conducted to determine effect of lablab green manure on population of soil microorganisms and establishment of common bean.

## 2. Materials and Methods

## 2.1. Description of the Study Site

The study was conducted in Koibem and Kapkerer located in Nandi South Sub-County in the North of Rift valley covering an area of 2,884Km<sup>2</sup> and extends to the north to latitude 0°34'N [17]. The area experiences two diverse seasons, the long rain seasons occurring from March to August and the short rain seasons from September to December. The area receives an average annual precipitation of 1200mm to 2000mm with mean yearly temperature ranging from 18-25°C and the soils are characterized by fine drained clay loamy soils [18].

## 2.2. Experimental Design and Layout

Lablab variety Rongai was planted during the long rains at a spacing of 45cm×30cm and at flowering the vegetation was harvested, cut and incorporated in soil at rate of one ton per ha. In each plots measuring 4m by 6m, 10kg of lablab green manure treatments was applied by evenly distributing cut portions over the whole plots, and in four furrows per plot filled with 2.5kg of chopped lablab vegetation. Diamonium phosphate fertilizer was applied at the rate of 75kg/ha<sup>-1</sup> in furrows [19] while control plots had no treatment applied. Bean varieties KK8 and GLP2 were planted in the plots at a spacing of 50cm×10cm by placing two seeds per hill immediately after incorporation of the lablab green manure with a seed rate of 50kg/ha. The treatments were arranged in a randomized complete block design with a split plot arrangement. The bean varieties constituted the main plots while the treatments comprised the subplots. Data was collected on crop emergence, incidence, and severity of root rot, plant stand, and yield. Soil samples was collected for isolation of soil microorganisms and for determining soil nutrient status

## 2.3. Sampling Soil for Microbial Analysis

Soil samples were collected before treatments application and at the second, fourth and sixth week after emergence to evaluate the changes in soil's chemical and biological properties [20]. A trowel was used to collect five samples (200g) of soil in each plot following a zigzag sampling procedure. The samples were mixed to form a composite sample of 1 kg which was kept in plastic bags and placed in the shade to prevent dehydration. The samples were divided into two sub samples for microbial and nutrient analysis.

#### 2.4. Analysis of Soil Chemical Characteristics

Soil samples collected was air dried and then broken by pounding with pestle and mortar [19]. The samples were then subjected to chemical and physical analyses to measure pH, available nitrogen, organic carbon, and available phosphorus. Soil pH, organic carbon was determined volumetric method according to Walkley and Black method as described [21]. Available nitrogen was determined by Kjeldahl method, phosphorus was determined by Brays method and Potassium was determined by Flame photometer [22].

# 2.5. Determination of Population of Microorganisms in the Soil

Both bacteria and fungi in the soil was isolated by taking from each soil sample one gram and dissolved in 10 ml sterile distilled water and shaken for 30 minutes. One milliliter of the soil suspension was transferred into 9 ml of sterile distilled water, shaken and the ten-fold dilution repeated up to a dilution of 10<sup>-3</sup> [23]. One milliliter of the third dilutions was plated on molten Potato Dextrose Agar and Nutrient Agar medium, cooled to 45°C [23]. The plates with Nutrient Agar were incubated for 24 hours while those on PDA were incubated for a week at room temperature after which the numbers of bacterial and fungal colonies were counted. The different fungal and bacterial colony types were identified based on colony color, growth type, colony reverse color and color of mycelia [24]. Population of each type of fungi and bacteria was determined following the formula  $CFU/g = Total number of colonies \times Dilution factor.$ 

The identification of bacterial isolates was carried out on

overnight cultures based on cultural and morphological characteristics while the different fungi isolated were sub cultured on potato dextrose agar medium for identification. Different fungal genera were identified based on the colony and cultural characteristics. Cultural characteristics used in identification were spore morphology, mycelia septation and macroconidial shape. *Fusarium* isolates was sub-cultured on synthetic nutrient agar [25] and on potato dextrose agar media. Cultures on synthetic nutrient agar were incubated under UV light to facilitate conidial sporulation while those on PDA were incubated at the room temperature for 14 days [26]

## 2.6. Determination of Emergence and Plant Stand

The emergence of common bean from the soil was assessed by counting the number of emerged plants one week after emergence while plant stand was determined by counting the number of surviving plants in each plot at the second, fourth and at sixth week after emergence.

#### 2.7. Assessment of Incidence and Area Under Disease Progress Curve of Root Rot

The incidence of root rot was determined by counting the number of seedlings with root rot symptoms in every plot at the second, fourth and sixth week after emergence [27]. Infected plants were identified based on symptoms like yellowing of leaves, wilting, stunted growth, brown discoloration on the roots and death [28]. Area under disease progress curve for disease incidence was calculated using the formula described by Muengula-Manyi *et al.*, [29]. AUDPC=  $\sum_{i=1}^{n} (Yi + Yi + 1)/2 (t2 - t1)$  where Yi is the incidence of disease at time I, Yi+1 is the disease incidence recorded at the time i+1, n, the number of registration on the incidence, and, days between the registration of Yi and Yi+1.

#### 2.8. Assessment of Yield and Yield Components

Yield attributes was determined by taking 10 samples of bean plants randomly from two central rows in each experimental unit at physiological maturity [30]. At harvest, plant biomass was determined by sampling from each plot ten plants that were completely dried in an oven at 50°C for a week and weighed then converted into kilogram per hectare. The total grain yield from each plot was weighed and converted into tons per hectare using the formula by Mwangi *et al.*, [2].

 $Yield = \frac{Field weight per plot (g) \times 1000 m2/ha}{Harvest area (m2) \times 1000 000 g/ton}$ 

#### 2.9. Data Analysis

Data was subjected to Analysis of variance using Genstat Inc. 15<sup>th</sup> edition 9 [31]. Means were separated by Fishers Protected LSD test. Correlation analysis was performed between soil microorganisms, disease incidence, and yield

#### 3. Results

#### **3.1. Soil Chemical Properties**

The chemical characteristics of soil were affected by incorporation of green manure in the soil (Table 1). The initial soil characterization of the study site indicated strong acidity in both sites with low percent organic carbon content. However, soil analysis done on samples taken from the farms six weeks after emergence showed that percentage organic matter in soil samples treated with green manures increased marginally with values ranging from 2.2% to 2.4% in Koibem while in Kapkerer the values ranged from 0.50 to 1.00, a similar trend due to green manure treatments can also be reported for soil pH, nitrogen, phosphorus in Kapkerer. The increases in soil organic carbon, and available nitrogen were highest for lablab treated plots.

	Koibem					Kapker	Kapkerer				
Treatments	pН	OC	Ν	Р	K	pН	OC	Ν	Р	K	
		%		Ppm	me%		%		ррт	me%	
Before incorporation	4.80	2.30	0.20	15.0	0.62	4.60	0.80	0.10	17.5	0.18	
Whole Incorporated	5.10	2.40	0.22	10.0	0.52	4.80	1.00	0.12	30.0	0.20	
Between rows	4.60	2.20	0.19	10.0	0.50	4.80	0.50	0.07	25.0	0.16	
No amendment	4.60	2.50	0.22	15.0	0.54	4.90	0.70	0.10	20.0	0.20	
DAP	4.67	2.10	0.18	15.0	0.50	4.69	0.88	0.11	25.0	0.16	

 Table 1. Soil chemical characteristics before and after lablab green manure incorporation.

DAP-Diamonium phosphate, N-nitrogen, P- phosphorus, K- potassium, OC- organic carbon, Me- milliequivalent, PPM-Parts per million

#### **3.2. Emergence and Plant Stand**

Significant differences (P  $\leq 0.05$ ) were observed between the different treatments with respect to emergence and plant stand in both seasons (Table 2). In both seasons, highest crop emergence was observed in Koibem than Kapkerer. Highest percentage crop emergence in both sites and in both seasons was recorded in DAP treated plots and in untreated plots while plots treated with lablab residues both in rows and in whole plots had the lowest percentage emergence in both seasons. However, there was increased crop emergence in the second season compared to the first (8%). In both sites and in both seasons, in the second, fourth and sixth week after emergence, plots without any treatment applied plots and plots treated with DAP had the highest stand count overtime when compared with plots incorporated with lablab green manure, however, in addition, there was no significant difference between the sites with respect to percentage stand count six weeks after emergence. There was a progressive to the week six. decline in plant stand across the sites from the second week

	Weeks after	emergence								
Treatments	Koibem				Kapkerer	Kapkerer				
	0	2	4	6	0	2	4	6		
2015 Short rains										
Lablab whole	55.4 <sub>ab</sub>	53.6 <sub>ab</sub>	49.3 <sub>ab</sub>	30.7 <sub>a</sub>	32.2 <sub>c</sub>	31.7 <sub>c</sub>	31.0 <sub>c</sub>	15.5 <sub>a</sub>		
Lablab in rows	50.3 <sub>ab</sub>	48.2 <sub>ab</sub>	44.2 <sub>abc</sub>	25.5 <sub>a</sub>	39.4 <sub>bc</sub>	39.2 <sub>bc</sub>	37.4 <sub>bc</sub>	17.0 <sub>a</sub>		
No amendment	63.2 <sub>a</sub>	62.2 <sub>a</sub>	56.8 <sub>a</sub>	32.7 <sub>a</sub>	60.0 <sub>a</sub>	60.0 <sub>a</sub>	56.6 <sub>a</sub>	24.6 <sub>a</sub>		
DAP	60.6 <sub>a</sub>	58.8 <sub>a</sub>	55.2 <sub>a</sub>	31.3 <sub>a</sub>	58.0 <sub>a</sub>	58.0 <sub>a</sub>	56.1 <sub>a</sub>	24.2 <sub>a</sub>		
Mean	57.4	55.7	51.4	30.1	47.4	47.2	45.3	20.3		
LSD ( $p \le 0.05$ )	10.6	11.1	11.1	17.2	10.6	11.1	11.1	17.2		
CV (%)	24.0	25.5	27.1	79.1	24.0	25.5	27.1	79.1		
2016 Short rains										
Lablab whole	46.3 <sub>c</sub>	41.9 <sub>d</sub>	31.5 <sub>f</sub>	19.9 <sub>e</sub>	41.5 <sub>c</sub>	39.6 <sub>d</sub>	37.1 <sub>f</sub>	27.1 <sub>de</sub>		
Lablab in rows	47.1 <sub>c</sub>	43.7 <sub>d</sub>	40.3 <sub>ef</sub>	26.5 <sub>de</sub>	48.4 <sub>c</sub>	44.7 <sub>d</sub>	43.7 <sub>def</sub>	31.9 <sub>cde</sub>		
No amendment	66.5 <sub>b</sub>	62.7 <sub>c</sub>	54.3 <sub>cde</sub>	32.1 <sub>cde</sub>	70.3 <sub>ab</sub>	65.5 <sub>bc</sub>	59.6 <sub>bcd</sub>	45.9 <sub>bc</sub>		
DAP	73.9 <sub>ab</sub>	69.9 <sub>abc</sub>	62.1 <sub>abc</sub>	38.9 <sub>bcd</sub>	72.0 <sub>ab</sub>	65.4 <sub>bc</sub>	63.7 <sub>abc</sub>	49.5 <sub>bc</sub>		
Mean	58.5	54.6	47.1	29.4	58.1	53.8	51.1	38.6		
LSD ( $p \le 0.05$ )	9.7	9.5	9.9	10.8	9.7	9.5	9.9	10.8		
CV (%)	18.9	19.7	22.5	34.2	18.9	19.7	22.5	34.2		

Table 2. Percentage plant stand at different sampling times after green manure incorporation.

DAP-Diamonium phosphate

Means within column followed by different letters are significantly different based on Fishers Protected LSD test ( $P \le 0.05$ ).

#### 3.3. Incidence and Severity of Root Rot

Significant differences ( $P \le 0.05$ ) were observed among the treatments in the two sites (Table 3). The highest root rot incidence was recorded in Kapkerer than in Koibem, however, overtime, there was reduction root rot incidence in Koibem while there was an increase in root rot incidence in Kapkerer. Highest root rot incidence was recorded in plots incorporated with lablab residues both in rows and in whole plots while the plots with not treatments applied had the least root rot incidence in both sites. Four weeks after emergence, root rot incidence in Kapkerer increased in all the treatments but plots incorporated with lablab green manure both in row

and in whole plots recorded the highest root rot incidences (32%). Six week after emergence, significant differences were observed between the treatments and there was decline in root rot incidences in all the treatments. In the second season of 2016, there were significant differences ( $P \le 0.05$ ) among the treatments within the two sites. Highest root rot incidence was recorded in Kapkerer than in Koibem. Addition of lablab residues in whole plots resulted in the highest mean root rot incidence in Kapkerer (23%) and Koibem (14%) while untreated plots recorded the least root rot incidence in both sites

Table 3. Percentage incidence of root rot at different sampling times after green manure incorporation.

Weeks after emergence						
Treatments	Koibem			Kapkerer		
Short rains 2015	2	4	6	2	4	6
Lablab whole	18.9 <sub>abc</sub>	20.1 <sub>abc</sub>	4.2 <sub>ab</sub>	30.4 <sub>a</sub>	31.9 <sub>a</sub>	6.3 <sub>a</sub>
Lablab in rows	17.4 <sub>bc</sub>	15.3 <sub>bc</sub>	4.1 <sub>ab</sub>	23.9 <sub>ab</sub>	27.1 <sub>ab</sub>	4.5 <sub>ab</sub>
No amendment	11.2 <sub>c</sub>	10.4 <sub>c</sub>	3.1 <sub>b</sub>	12.0 <sub>bc</sub>	15.5 <sub>bc</sub>	5.7 <sub>ab</sub>
DAP	11.4 <sub>c</sub>	11.1 <sub>c</sub>	4.0 <sub>ab</sub>	14.1 <sub>bc</sub>	17.7 <sub>bc</sub>	4.8 <sub>ab</sub>
Mean	14.7	14.2	3.9	20.1	22.2	5.3
LSD ( $p \le 0.05$ )	7.4	7.6	1.9	7.4	7.6	1.9
CV (%)	56.9	53.6	53.1	56.9	53.6	53.1
Short rains 2016						
Lablab whole	19.6 <sub>bc</sub>	13.7 <sub>de</sub>	11.8 <sub>c</sub>	31.2 <sub>a</sub>	34.4 <sub>a</sub>	37.7 <sub>a</sub>
Lablab in rows	17.4 <sub>bc</sub>	15.2 <sub>cde</sub>	12.5 <sub>c</sub>	24.7 <sub>ab</sub>	29.8 <sub>ab</sub>	30.9 <sub>ab</sub>
No amendment	11.2 <sub>c</sub>	11.2 <sub>de</sub>	10.7 <sub>c</sub>	15.8 <sub>c</sub>	19.7 <sub>cd</sub>	22.4 <sub>b</sub>
DAP	11.4 <sub>c</sub>	11.4 <sub>de</sub>	11.1 <sub>c</sub>	19.3 <sub>bc</sub>	23.4 <sub>bc</sub>	27.3 <sub>b</sub>
Mean	14.9	12.9	11.5	22.8	26.8	29.6
LSD ( $p \le 0.05$ )	7.3	5.7	5.8	7.3	5.7	5.8
CV (%)	49.1	37.8	36.5	49.1	37.8	36.5

DAP-Diamonium phosphate

Means within column followed by different letters are significantly different based on Fishers Protected LSD test ( $P \le 0.05$ ).

On area under diseases progress curve, there were significant differences (p  $\leq$  0.05) among the treatments in both seasons (Figure 1). Largest area under diseases progress curve was observed in the 2016 short rains than in the 2015 short rain season. However, among the treatments, plots treated with lablab green manure had the largest area under diseases progress

curve (827.3) while those in untreated plots and in DAP treated plots had the least AUDPC for root rots of common bean.



DAP-Diamonium phosphate

Figure 1. Area under disease progress curve of common bean root rot on KK8 and GLP2 bean varieties.

#### 3.4. Population of Root Rot Pathogens Isolated from the Soil

The relative abundance of the *Fusarium* spp. isolates recovered was quantified (Table 4) and species including *F. solani, F. oxysporum* were dominant accounting for more than 80% of root rot pathogens isolated from the soil. *Pythium* species albeit in small proportions was also isolated from the soil samples. The populations of these pathogens were significantly affected by green manure treatments, in the first season, soil samples collected from Kapkerer site had the highest population density of root rot pathogens compared to those from Koibem, however, in the second season, the population of root rot pathogens from soil samples collected from Koibem were higher than those collected from Kapkerer. In both sites and in both seasons the densities of both *Fusarium solani* and *Fusarium oxysporum* were significantly higher in plots treated with lablab green manure compared with other treatments

Tuestments	Koibem			Kapkerer	Kapkerer				
1 reatments	F. solani	F. oxysporum	Pythium	F. solani	F. oxysporum	Pythium			
2015 short rains									
Lablab whole	2.7 <sub>b</sub>	3.2 <sub>a</sub>	0.61 <sub>b</sub>	4.3 <sub>a</sub>	4.7 <sub>a</sub>	0.89a			
Lablab in rows	3.8 <sub>a</sub>	3.3 <sub>a</sub>	0.79 <sub>a</sub>	4.7 <sub>a</sub>	4.0 <sub>b</sub>	0.60 <sub>b</sub>			
No amendment	2.6 <sub>b</sub>	3.1 <sub>a</sub>	0.71 <sub>a</sub>	3.4 <sub>b</sub>	3.9 <sub>bc</sub>	0.67 <sub>b</sub>			
DAP	2.7 <sub>b</sub>	2.8 <sub>b</sub>	0.54 <sub>b</sub>	3.1 <sub>b</sub>	3.3 <sub>c</sub>	0.83 <sub>a</sub>			
Mean	2.9	3.1	0.7	3.9	4.0	0.7			
LSD (p≤0.05)	0.6	0.3	0.1	0.8	0.7	0.2			
2016 short rains									
Lablab whole	2.3 <sub>a</sub>	1.8 <sub>a</sub>	0.75 <sub>b</sub>	2.7 <sub>a</sub>	2.2 <sub>a</sub>	1.02 <sub>a</sub>			
Lablab in rows	2.5 <sub>a</sub>	1.8 <sub>a</sub>	1.30 <sub>a</sub>	2.5 <sub>a</sub>	1.8 <sub>a</sub>	1.03 <sub>a</sub>			
No amendment	1.9 <sub>b</sub>	1.2 <sub>b</sub>	1.00 <sub>ab</sub>	1.6 <sub>b</sub>	1.4 <sub>b</sub>	0.90 <sub>a</sub>			
DAP	1.9 <sub>b</sub>	1.2 <sub>b</sub>	0.85 <sub>b</sub>	1.8 <sub>b</sub>	1.5 <sub>b</sub>	0.71 <sub>b</sub>			
Mean	2.2	1.5	0.97	2.2	1.7	0.92			
LSD ( $p \le 0.05$ )	0.3	0.44	0.32	0.73	0.4	0.21			

**Table 4.** Population  $(10^4)$  of root rot pathogens isolated from soils incorporated with lablab green manure.

DAP-Diamonium phosphate

Means within column followed by different letters are significantly different based on Fishers Protected LSD test ( $P \le 0.05$ ).

#### 3.5. Population of Saprophytic Microorganisms Isolated from the Soil

The results obtained from the experiments indicate that

application of lablab green manure had significant effect on

quantitative composition of beneficial saprophytes in the soil (Table 5). Species including *Aspergillus*, *Trichoderma*, and *Penicillium* were dominant. In the first season, it was observed that samples from Kapkerer site had the highest

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population density of saprophytes than Koibem. In both sites and in both seasons, the densities of all the saprophytes were higher in plots treated with lablab green manure followed by plots treated with diamonium phosphate fertilizer and in plot without any treatment. The population of bacteria did not vary between the sites and in both seasons ( $p \le 0.05$ ). In the first season in Koibem, plots

with no treatment application had the highest bacterial density while plots treated with lablab had the lowest bacterial density while, there was no significant difference between the treatments in Kapkerer. In the second season, in both sites there was no significant difference ( $p \le 0.05$ ) between the treatments.

 Table 5. Population  $(10^4)$  of saprophytic fungi and bacteria isolated from plots incorporation with lablab green manure.

Treatments	Koibem				Kapkerer			
	Asper	Tricho	Pen	Bacteria (10 <sup>6</sup> )	Asper	Tricho	Pen	Bacteria (10 <sup>6</sup> )
2015 short rains								
Lablab whole	1.9 <sub>a</sub>	1.1 <sub>ab</sub>	1.4 <sub>a</sub>	23.9 <sub>ab</sub>	0.9 <sub>bc</sub>	0.9 <sub>b</sub>	1.5 <sub>b</sub>	13.4 <sub>b</sub>
Lablab in rows	1.1 <sub>b</sub>	1.3 <sub>a</sub>	0.9 <sub>b</sub>	27.8 <sub>ab</sub>	1.1 <sub>a</sub>	1.4 <sub>a</sub>	1.7 <sub>b</sub>	16.6 <sub>b</sub>
No amendment	1.4 <sub>b</sub>	1.4 <sub>a</sub>	1.1 <sub>ab</sub>	41.4 <sub>a</sub>	1.2 <sub>a</sub>	0.8 <sub>b</sub>	2.7 <sub>a</sub>	12.3 <sub>b</sub>
DAP	1.2 <sub>b</sub>	0.7 <sub>bc</sub>	1.1 <sub>ab</sub>	25.3 <sub>ab</sub>	0.8 <sub>c</sub>	0.5 <sub>c</sub>	1.4 <sub>b</sub>	12.0 <sub>b</sub>
Mean	1.4	1.2	1.1	29.6	1.0	0.9	1.8	13.6
LSD (p≤0.05)	0.4	0.51	0.35	12.3	0.17	0.3	0.79	12.3
CV (%)	0.13	0.09	0.21	8.0	0.03	0.14	0.36	4.4
2016 short rains								
Lablab whole	1.7 <sub>a</sub>	0.7 <sub>ab</sub>	2.1 <sub>ab</sub>	18.9 <sub>a</sub>	0.8 <sub>b</sub>	0.5 <sub>b</sub>	0.9 <sub>b</sub>	15.0 <sub>a</sub>
Lablab in rows	0.7 <sub>b</sub>	0.9 <sub>a</sub>	0.9 <sub>b</sub>	13.2 <sub>a</sub>	1.4 <sub>a</sub>	1.2 <sub>a</sub>	2.6 <sub>a</sub>	18.9 <sub>a</sub>
No amendment	0.9 <sub>b</sub>	0.9 <sub>a</sub>	2.6 <sub>a</sub>	19.2 <sub>a</sub>	1.3 <sub>a</sub>	0.7 <sub>b</sub>	2.3 <sub>a</sub>	18.6 <sub>a</sub>
DAP	0.8 <sub>b</sub>	0.5 <sub>b</sub>	1.9 <sub>ab</sub>	20.0 <sub>a</sub>	0.7 <sub>b</sub>	0.5 <sub>b</sub>	1.8 <sub>a</sub>	18.3 <sub>a</sub>
Mean	1.0	0.8	1.9	17.8	1.1	0.7	1.9	17.7
LSD ( $p \le 0.05$ )	0.5	0.2	0.8	99.3	0.3	0.3	0.8	99.3
CV (%)	0.21	0.03	0.51	9.7	0.12	0.11	0.55	3.3

DAP-Diamonium phosphate

Means within column followed by different letters are significantly different based on Fishers Protected LSD test ( $P \le 0.05$ ). Asper- Aspergillus, Pen-Penicillium, Tricho-Trichoderma

#### 3.6. Grain and Biomass Yield of Common Beans

There was significant ( $p \le 0.05$ ) difference in biomass yield of common beans in Koibem (Table 6). In the first season, plots treated with DAP resulted in high biomass and grain yield across the two sites, Koibem (343kg/ha<sup>-1</sup>, 3.4t ha<sup>-1</sup>) while in Kapkerer untreated plots had the highest yield

(267.0kg/ha<sup>-1</sup>, 2.4t ha<sup>-1</sup>) while lablab incorporated plots consistently gave the lowest yield in the first season and in the second season averaging (2t ha<sup>-1</sup>). Unfertilized controlled plots performed better than the plots that were incorporated with lablab green manure. In general DAP applied plots resulted in improved common beans yield as compared to other treatments.

Table 6. Effect of lablab green manure on common bean biomass and grain yield.

Treatments	Koibem		Kapkerer	
2015 Short rains	Plant Biomass (Kg/ha)	Grain Yield (t/ha)	Plant Biomass (Kg/ha)	Grain Yield (t/ha)
Lablab whole plots	243.5 <sub>c</sub>	2.4 <sub>a</sub>	276.5 <sub>a</sub>	2.2 <sub>a</sub>
Lablab Between rows	257.2 <sub>bc</sub>	2.7 <sub>a</sub>	267.9 <sub>a</sub>	2.0 <sub>a</sub>
No amendment	345.4 <sub>a</sub>	2.7 <sub>a</sub>	265.7 <sub>a</sub>	2.4 <sub>a</sub>
DAP	343.6 <sub>ab</sub>	3.4 <sub>a</sub>	260.5 <sub>a</sub>	1.9 <sub>a</sub>
Mean	297.4	2.8	267.7	2.1
LSD ( $p \le 0.05$ )	117.4	1.4	170.8	1.7
CV (%)	25.7	27.9	34.9	33.8
Short rains 2016				
Lablab whole plots	56.7 <sub>e</sub>	0.21 <sub>e</sub>	160.7 <sub>cde</sub>	0.5 <sub>cde</sub>
Lablab between rows	117.2 <sub>de</sub>	0.35 <sub>de</sub>	198.9 <sub>bcde</sub>	0.6 <sub>bcde</sub>
No amendment	161.5 <sub>cde</sub>	0.6 <sub>bcde</sub>	253.9 <sub>bcd</sub>	1.0 <sub>bcd</sub>
DAP	151.8 <sub>cde</sub>	0.7 <sub>bcde</sub>	345.8 <sub>ab</sub>	1.2 <sub>ab</sub>
Mean	158.1	0.575	288.8	1.04
LSD ( $p \le 0.05$ )	108.9	0.43	108.9	0.43
CV (%)	60.2	67.2	60.2	67.2

DAP-Diamonium phosphate

Means within column followed by different letters are significantly different based on Fishers Protected LSD test ( $P \le 0.05$ ).

#### 3.7. Relationships Among Root Rot Pathogens, Saprophytes, Root Rot Incidence and Yield

According to correlation analysis, the strongest negative relation was displayed between rot pathogens *F. oxysporum* and *Pythium* (-0.567, -0.661,  $P \le 0.05$ ) to crop emergence and plant stand (-0.294, -0.163  $P \le 0.05$ ) respectively. Isolation frequency of root rot pathogens from soil was negatively correlated to beneficial saprophytic fungi isolated from the soil. There was strong negative correlation between

*F. solani, F. oxysporum* and *Pythium* (-0.578, -0.626, -0.606 P  $\leq$  0.05) respectively to yield (Table 7), however, the saprophytic fungi had significantly positive relation with yield. The crop emergence was positively correlated to yield (+0.776, P  $\leq$  0.05) but was negatively correlated with to root rot incidence (-0.547, P  $\leq$  0.05). Root rot incidence correlated negatively with grain yield and beneficial saprophytic (-0.687, P  $\leq$  0.05) but was positively associated with the population of *F. solani, F. oxysporum* and *Pythium* 

Table 7. Correlation coefficients among root rot pathogens, saprophytes, emergence, disease incidence, and yield.

	F. 0	<i>F. s</i>	Pyth	Trich	Pen	Asper	Emerg	P. S	Incide	Yield (t/ha)
F. oxysporum	-									
F. solani	0.398	-								
Pythium	0.186	-0.009	-							
Trichoderma	0.056	-0.157	0.080	-						
Penicillium	0.099	-0.138	0.139	0.400	-					
Aspergillus	-0.081	-0.316	0.002	0.141	0.009	-				
Emergence	-0.567*	0.137	-0.661*	-0.202	-0.240	-0.053	-			
Plant stand	-0.294	0.016	-0.163	-0.182	-0.193	-0.075	0.493	-		
Incidence	0.524*	0.701*	0.403	0.205	-0.014	0.069	-0.547	-0.286	-	
Yield (t/ha)	-0.578*	-0.626*	-0.606*	0.824	0.842	0.275	0.776*	0.557	-0.687*	-

\* Significantly correlated

### 4. Discussion

Organic Matter (OM), pH, available nitrogen, available phosphorus and exchangeable potassium of the plots treated with organic amendment marginally increased. This is due to the addition of organic amendments that are the source of organic carbon and nitrogen to the soils. The changes in soil properties are associated with the type of soil amendment applied [32]. The results herein concur with those reported by Zeid et al., [33] who found that organic materials applied alone or in combination with inorganic fertilizers increase soil fertility. The soils where green manure was incorporated had higher nitrogen concentrations than the untreated plots. This is because of higher nitrogen levels in lablab that result in increased amounts of N fixed by legume [34]. Leguminous green manures provide large amount of nitrogen available in soils by the decomposition of biomass [35]. Soil organic carbon is a source for plant nutrients in soils and maintains soil silt thereby aiding infiltration of air and water [36]. The result of the soil chemical parameters thus indicated that the treatments applied had effect on the chemical parameters of the soil albeit in small quantities

The highest germination percentage was observed in plots treated with DAP while lablab treated soils consistently had the lowest germination percentage, these results are comparable to findings of the preceding work by Muthomi *et al.*, [27] that common bean emergence and establishment are sensitive to green manure from lablab. Our results also agree with findings by Bonanomi *et al.*, [37] who reported germination inhibition of about 48%, however, the response varied according to litter type. Wuest and Skirvin, [38] reported various substances extracted from fresh residues that inhibited growth of plants in the laboratory. These substances

can inhibit microbial growth in the soil and also make plant roots vulnerable to root infection. The present study indicated specificity of action of the residues and the concentrations effects of the inhibitors on common beans emergence. Wuest and Skirvin, [38] noted that the decrease in crop emergence could be as a result of phytotoxins since there was increased germination after washing the soil with water. Green crop residues during decomposition may result in production of phytotoxic products that may limit seedling germination and establishment. The initial stage of decomposition involves breakdown of plant tissue by microorganisms and subsequent release of contents of the cell with oxidized functional group while during late stages there is an increase of phenolic functional groups indicating degradation of lignin in the maturing compost [37]. Chemical nature and toxicity of inhibitors derived from undecomposed plant residues are different. The residues contain water soluble phytotoxins which are responsible for depressed germination and seedling establishment [37]. The growth inhibitors from decomposing plant residues are most concentrated in the soil close to particles of decomposing plant materials, roots of common beans are sensitive to these chemicals and showed symptoms of damage which resulted in stunted growth of seedlings. Phytotoxicity was severe in the initial stages of decomposition since toxicity of green manure develops relatively early in the decomposition process. When lablab green manure was applied in rows, inhibitory substances produced during decomposition were also unevenly distributed and relatively uniform damage was observed in beans planted in the same rows as lablab green manure. Soils modified by addition of green manure preconditions plant roots to attacks by root rot pathogens and the phytotoxins produced during decomposition conditions roots for pathogen

attack. This may explain the role of lablab green manure as physical impediment to emerging seedlings and the etiology of pathogen attack of young seedlings.

Population of Fusarium root rot pathogens in lablab treated plots was higher in the second week after treatment applications. Interactions of green manure with fungal pathogens like Fusarium may be involved in suppression of germination of common beans. Bonanomi et al., [37] showed that undecomposed plant residues promote fungal growth, although the effect on microbial response can vary depending on the residue type applied in the soil. However, as decomposition proceeds, plant residues become more suitable for plant growth while bacteria and fungi are inhibited. Addition of green manure improves speedy multiplication of microorganisms in the soil [39] resulting in increased metabolism which causes high consumption of oxygen in soil resulting in increased carbon dioxide production which may retard common bean germination [40]. Large number of organisms in soil may also result in possible accumulation of toxic substances to germination. These waste products are less decomposable than the original plant material.

The results obtained from both seasons indicate that incorporation of lablab green manure diversified the numerical and qualitative composition of soil fungi. Pathogenic Fusarium oxysporum, Fusarium solani and Pythium spp were found to dominate plots treated with lablab green manure. There population increased two weeks after treatment applications and at the same time reduced the population of saprophytic fungi. This finding is consistent with Abawi and Widmer [41] who reported an increase in pathogenic fungi with organic fertilizer application. Fresh green manure results in rapid increase in soil microbial biomass, of which fungi are commonly the largest component [42]. Pathogenic Fusarium and Pythium species cause reduction in seed germination potential [43]. Symptoms associated with undecomposed plant residue were evident immediately after planting but overtime the plant partially recovered from wilting. Bonanomi et al., [44] reported increased symptoms such as stunting, yellowing and wilting following green manure application. Decomposition of lablab residue shows their negative effects as soon as they come into contact with plant roots because they interfere with water uptake [45]. The addition of green manure provides significant inputs of organic carbon, which increases fungal population [46]. Fresh green manure and the amount returned to the soil improve microbial activity [47] and provide significant inputs of organic carbon, which increases both fungal and bacterial populations [46]. The fungi from the control plots help in estimating and identifying the indigenous fungal population and diversity of the study sites.

Antagonistic fungi isolated in small proportions were *Trichoderma, Penicilium* and *Aspergillus.* High concentrations of nutrients in fresh green manure residues inhibit the production of enzymes required for parasitism by biocontrol agents such as *Trichoderma* spp. [48]. As a consequence, the weakened biological barrier due low

population of saprophytic fungi could be the cause of intensive symptoms of root rot disease. The environment in which green manures is applied may contain high concentrations of salt, ammonium salt and low oxygen which is unsuitable for growth and multiplication of antagonistic microorganisms [49]. Higher levels of soil infestation with root rot pathogens result in increased disease potential as shown in plots with lablab green manure incorporated either in rows or fully on the plot. This result conforms to findings by Stone and Hansen [50] who reported higher levels of soil infestation following plant residue incorporation and therefore increases in disease pressure. The fungal response to available plant materials in the soil suggests the need for great care in managing green manure and plant debris incorporation [16]. In order to exploit the richness in moisture, nutrient and physical soil characteristics, there should be great care in organic farms where crops are planted immediately after green manure incorporation. The same findings were reported by Ambrosano et al., [51], however, the researchers were working on corn as test plant and after the initial period the productivity of corn was comparable to the control.

The addition of soil amendments resulted in either reduced population or maintained the initial population of antagonists but appeared to support the population of pathogenic fungi. These changes were observed in the initial stages of decomposition in all the farms regardless of previous history of the farm in which the experiment was conducted. Green manure stimulates biological and microbial activities thus speed up the breakdown of organic substances [52]. Our results show that the soil amendments used had no suppressive effects on pathogenic microbes like Fusarium species and Pythium. The negative effect of green manure has been verified with the use of red clover green manure where there was increased incidence of disease in wild mustard seeds [53]. However, other studies by Wigins and Kinkel, [54] suggest that green manure treatments may help in disease control by activating pathogen, by releasing phytotoxins made during product storage or by subsequent microbial decomposition

In intensively cultivated soils like the ones where the experiment was set up, where saprophytic pathogens have been increased by earlier soil management practices, the ploughing in of organic debris like green manures enhances the population of the pathogens and in so doing increasing root rot incidence and severity in subsequent crops. Exhaustive cropping system leads to poor soil fertility with low organic matter in the soil accompanied by increased pathogenic population in the soil [42]. The response of the pathogens and the lack of suppression of the same pathogens reveal that the pathogens may pose potential problems during the first two weeks after green manuring in the soil thus result in poor crop germination and establishment.

Results observed shows that beans planted in plots treated with DAP fertilizer and in control plots outperformed plots treated with green manure. The differences observed between the treatments may be as a result of soil nutrient content, type and nature of the microorganism present in the soil [55]. Yields decrease with increase in incidence and severity of soilborne diseases [56]. Yield increases in organic production systems, however, this was not the case in this study. To record high yields in organically amended fields, organic productions systems require 3-5 years of manure application [57] to be more productive due to beneficial effects on soil properties of long term soil organic amendments applications [58]. This explains the reason our results do not support the high yield theory when it comes to green manure application since it was not long term and plants were planted immediately after treatments applications. In this research, correlation analysis identified the association between root rot pathogens, emergence, plant stand and crop vield as an important descriptor due to their evident relationships. At the same time, there was a negative interrelationship between the root rot pathogens and beneficial fungi. Such at one point high population of pathogenic fungi in the soil may affect crop emergence thus reduction in establishment and poor yield.

## 5. Conclusion

The results demonstrate that lablab green manure enhances chemical attributes and significantly impacts on diversity, composition and structure of soil microbial communities but results in poor crop emergence. Given the complex nature of the interactions among residues in soil, microbes, plants as well as soil conditions, it becomes difficult to demonstrate that buried lablab residues reduce crop emergence and vigour. However, understanding these effects is important to manage green manure hence focusing on the balance between negative and positive effects. The research underlines the need to apply green manure at the right time without compromising crop establishment thus accruing economic benefits

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