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Effect of Time after Incorporation of Lablab Green Manure on Root Rot Pathogens and Establishment of Common Bean (*Phaseolus vulgaris* L.)

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Abstract Green manure incorporation is important for restoration of soil quality, particularly buildup of organic matter and supply of nutrients to plants. However, undecomposed plant residues reduce crop establishment and plant stand. Therefore, there is need to determine suitable time for green manure incorporation before planting. The effect of time after incorporation of lablab green manure on soilborne pathogens and bean crop establishment was evaluated by incorporating 12t/ha of lablab green manure at planting and at 7, 14, and 28 days before planting. Soil samples were collected before and after incorporation of green manure at planting, and at two, four and six weeks after planting. Data was collected on crop emergence, plant stand, yield, incidence and severity of root rot, and population of root rot pathogens. Incorporation of lablab residues 28 days before planting resulted in 21% improvement in germination, with corresponding reduction in root rot incidence and severity of 8% and 36%, respectively, compared to plots incorporated with green manure at planting. Plots incorporated with lablab green manure earlier before planting had reduced population of root rot pathogens, while those incorporation at planting excited the population of root rot pathogens and also had up to 71% reduction in grain yield compared to plots where lablab residue was incorporated 28 days before planting. The results of the study showed that a period of 28 days between Lablab green manure incorporation and planting is necessary to allow for proper decomposition, resulting in a reduction in root rot incidence and an increase in grain yield.

Keywords: *soil health, green manures, Lablab purpureus, soil borne pathogens*

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1. Introduction

Common bean (*Phaseolus vulgaris* L.) is known as staple legume [1] and low cost protein source consumed in many developing countries where protein malnutrition is prevalent [2]. However, productivity is very low in Africa and Latin America [3]. Decline in soil fertility is a major constraint to bean production and farmers usually apply fertilizer below recommended rates along with agricultural practices that lead to nutrient depletion [4]. Application of green manure is considered a sustainable option for the supply of nitrogen for organic production systems [5] since the release of nutrients from these types of manure is slower and therefore contributes to better production system sustainability [6]. Green manure may be obtained from legumes and perennial woody trees [7]. Plant residues are important component of the soil and they can either be living, dying, or dead with enormous chemical diversity, and are eventually decomposed through the action of biotic and abiotic agents [8]. The residue decomposition process is influenced by various factors including environment, decomposer community and residue quality [9]. Soil fauna positively influences decomposition rates and mobilizes nutrients from organic residues [10].

The process of decomposition involves numerous multifaceted interactions and transformations [11] such that at one point, the soil environment could contain a diverse chemical compounds which may have positive or negative effects on all phases of plant development. Schroeder *et al.* [12] working with cowpea residue found that when the residues were incorporated before broccoli crop, higher seedling mortality occurred. The researchers concluded that undecomposed plant residues incorporated into the soil before planting had deleterious effects including release of phytotoxic substances, and enhancement of pathogenic fungi [13]. There effects on seedling growth and disease incidence

depend on maturity of the tissues, carbon: nitrogen ratio and time elapsed between incorporation and planting [14]. Incorporation of green manure is immediately followed by an explosion of microbial activity and therefore if a susceptible crop is planted immediately they become invaded by these pathogenic microorganisms. Phytotoxic effects from decomposing plant residues increase within the first days after incorporation then decrease. Thus the effects on the seedling establishment may be through competition for nutrients or through injury by the substances produced during decomposition or indirect through stimulation of pathogenic fungi [13]. Residue degradation depend on lignin and cellulose content and their carbon: nitrogen ratio, which is crop dependent, but the environment and soil conditions also play a role [14]. The injury to roots of seedling is mainly as a result of seedling root coming into contact with the immediate vicinity of the decomposing residues. Even with the importance of green manure, there is little information on how soon crops can be safely planted after application to achieve good establishment, especially common beans. Therefore, the objective of this study was to investigate effect of time after incorporation of lablab green manure on population of root rot pathogens and establishment of common bean.

2. Materials and Methods

2.1. Description of the Study Site

The study was conducted in Koibem and Kapkerer in Nandi South Sub-County located in western Kenya, within latitudes 0° and 0°34'' North and longitudes 34°44'' and 35°25'' East, at an elevation of 1850-2040m above sea level [15]. These areas receive an annual precipitation of about 1200mm to 2000mm with mean annual temperature of about 25°C with soils that are well drained clay-loams, classified as nitosols [16].

2.2. Experimental Design and Layout

Lablab variety Rongai was planted in plots measuring 4m by 6m at a spacing of 45cm by 30cm and at flowering stage the vegetation was harvested, chopped into tiny portions, weighed and incorporated at the rate of 12 tons/ha at 28, 14, 7 days before and at planting in the different plots. Common bean varieties KK8 and GLP2 were planted at a spacing of 50 by 10cm in each plot. The plots were separated by 1m paths and the treatments were organized in a randomized complete block design with a split plot arrangement where bean varieties comprised the main plots while incorporation times were the subplots. The study was carried out over two short growing seasons of 2016 and 2017. Data on crop emergence was collected one week after planting while, incidence and severity of root rot, and plant stand, were collected after every two weeks until flowering. Yield data was collected at maturity. Soil samples were collected for isolation of soil microorganisms at planting, followed by three other samplings at an interval of two weeks.

2.3. Isolation of Microorganisms from the Soil

Fungal populations in the soil were isolated by taking one gram of soil from each sample and dissolving in 10 ml sterile distilled water and thoroughly shaking 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 10^6 [17]. About 1ml of the third dilutions was plated on molten potato dextrose agar and nutrient agar medium, cooled to 45°C [17]. The plates with potato dextrose agar were incubated for seven days at 25°C for fungal growth after which numbers of fungal colonies were counted. The different fungal colony types were identified based on colony colour, growth type, colony reverse colour and colour of mycelia [18]. Population of each type of fungi was determined by multiplying the number of colonies by the dilution factor.

$$\text{CFU / g} = \text{Total number of colonies} \times \text{Dilution factor} \quad (1)$$

2.4. Determination of Emergence and Plant Stand

Emergence was determined one week after planting by counting the number of emerged plants while plant stand was determined by counting the number of surviving plants in each plot at the second, fourth and sixth week after emergence [19].

2.5. Determination of Incidence and Severity of Root Rot

Incidence of root rot was determined by counting the number of bean seedlings showing root rot symptoms per plot at the second, fourth and sixth week after crop emergence, while root rot severity was determined four weeks after emergence based on a scale of 0-5 [20] where 0-no root discoloration, 1- 1~25% root discoloration, 2-26~50% root discoloration, 3- 51~75% root discoloration, 4-up to 76% root discoloration, 5- completely dead plants. Area under disease progress curve for disease incidence was calculated using the formula described by [21].

$$\text{AUDPC} = \sum_{i=1}^{n} (Yi + Yi+1) / 2(t2 - t1) \quad (2)$$

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.

2.6. Determination of Yield and Yield Attributes

Yield and yield attributes of beans were determined by taking 10 samples of bean plants randomly from two central rows in each experimental unit at physiological maturity [22]. Plant biomass was determined by sampling ten plants from each plot, and separated into root and shoots. Roots were carefully separated from rhizospheric

soil by washing. Both shoots and roots were dried at 50°C for seven days to determine dry weight. The total yield from each plot was weighed and converted into tons per hectare using the formula by Mwangi *et al.*, [23].

$$\text{Yield} = \frac{\text{Field weight per plot(g)} \times 10000\text{m}^2 / \text{ha}}{\text{Harvest area(m}^2) \times 1000000\text{g} / \text{ton}}. \quad (3)$$

2.6. Data Analysis

The collected data was subjected to analysis of variance using Genstat (Version 15) computer software [24]. Mean separation was done by protected Fisher's Least-significant difference test (LSD) at $p \leq 0.05$.

3. Results

3.1. Emergence and Plant Establishment

The different lablab green manure incorporation times had significant ($P \leq 0.05$) effect on germination. The results shows that in the first short rain season of 2016 and at both the sites, plots incorporated at planting had the lowest percentage germination (84%) while the treatments incorporated before planting recorded higher germination percentages. However, there was decline in plant stand overtime. The results show that in the second short rain season of 2017, plots incorporated with lablab green manure at planting and those planted seven days after incorporation recorded considerably low germination percentages of 57% and 59%, respectively, while other incorporation times had higher emergence percentages. The greatest decline in plant stand across the two sites was observed in 2017. The common bean emergence was in the order of 28 > 14 > 7 > 0 days incorporation after planting. Plant stand declined markedly to low levels in Kapkerer and Koibem over the entire sampling period (Table 1).

3.2. Root Rot Incidence and Area under Disease Progress Curve

The different green manure incorporation times had significant effect on root rot incidence of beans. In the first season, there were significant ($p \leq 0.05$) differences between incorporation times, and higher root rot incidences were recorded in plots incorporated with green manure at planting in both sites (Table 2). However, two weeks after emergence, the incidence of root rots reduced at both sites but greatly increased in plots incorporated at planting at Koibem by 35% and Kapkerer 27% respectively. The incidence of root rot at Kapkerer continued to increase while at Koibem the incidence reduced six weeks after emergence. The results show that high root rot incidences were recorded in the second season which was characterized by long and heavy rainfall. In the second season, plots incorporated with lablab green manure at planting had the highest root rot incidence at both sites, while plots incorporated 14 and 28 days earlier before planting beans had the least incidence of root rot incidence. In all plots, incidence increased four weeks after emergence but reduced in the sixth week after emergence.

There were significant differences ($p \leq 0.05$) among the treatments and between the varieties in both seasons for area under disease progress curve (Table 3). The AUDPC varied from 211 to 957 for KK8 and 367 to 969 for GLP2. Largest area under diseases progress curve was recorded in the second season of 2017 compared to the first season of 2016 at both sites. In both seasons and sites, the plots incorporated at planting had the largest area under disease progress curve while those incorporated 28 days before planting had the least area under disease progress curve. The two varieties showed different levels of susceptibility with GLP2 variety being the most susceptible to root rots and had the largest area under disease progress curve in both seasons in plots incorporated at planting while variety KK8 had the least area under disease progress curve.

Table 1. Percentage plant stand of common bean at different sampling times after incorporation of lablab green manure

Days After incorporation	Weeks after emergence							
	Koibem				Kapkerer			
	0	2	4	6	0	2	4	6
2016 Short rain								
28 days	99.8_a	96.5_a	94.9_a	93.8_a	99.8_a	81.5_a	51.4_a	50.7_a
14 days	99.6_a	94.5_a	92.7_a	91.8_a	100.0_a	69.5_b	35.0_{ab}	34.4_b
7 days	96.4_a	92.8_a	90.7_a	89.8_a	100.0_a	69.7_b	50.4_a	49.7_a
0 days	82.1_b	62.6_b	59.5_b	57.9_b	87.2_b	70.3_b	39.6_{ab}	38.8_b
Mean	94.5	86.6	84.4	83.3	96.9	72.7	44.1	43.4
LSD ($p \leq 0.05$)	5.9	10.4	16.9	16.8	5.9	10.4	16.9	16.8
CV (%)	3.8	11.0	17.5	17.6	3.8	11.0	17.5	17.6
2017 Short rain								
28 days	84.1_a	80.6_a	61.8_a	30.4_a	78.7_a	74.0_a	52.8_a	32.9_a
14 days	84.2_a	78.3_a	47.8_b	24.3_a	75.7_a	68.5_{ab}	55.3_a	26.4_a
7 days	57.4_b	48.7_b	44.7_b	21.5_a	59.0_b	55.6_b	50.3_a	25.3_a
0 days	59.3_b	54.7_b	39.3_{bc}	28.4_a	46.3_b	43.2_b	40.8_{ab}	24.0_a
Mean	71.3	65.6	48.4	26.2	64.9	60.3	49.8	27.2
LSD ($p \leq 0.05$)	17.4	17.3	15.4	18.2	17.4	17.3	15.4	18.2
CV (%)	15.1	14.8	10.5	31.1	15.1	14.8	10.5	31.1

Values followed by the same letter within the same row are not significantly different between the treatments using Fishers Protected LSD test ($P \leq 0.05$).

Table 2. Percentage root rot incidence at different sampling times after green manure incorporation

Days after incorporation	Weeks after emergence					
	Koibem			Kapkerer		
	2	4	6	2	4	6
2016 Short rains						
28 days	0.9_b	6.8_b	10.6_b	0.5_a	12.6_b	28.4_a
14 days	0.89_b	5.2_b	4.3_b	0.4_a	12.8_b	40.1_a
7 days	1.4_b	4.9_b	7.6_b	0.9_a	21.5_{ab}	31.6_a
0 days	4.9_a	35.6_a	29.2_a	1.3_a	27.1_a	77.4_a
Mean	2.0	13.1	12.9	0.8	18.5	44.4
LSD ($p \leq 0.05$)	3.2	9.6	7.8	1.1	12.8	88.9
CV (%)	49.0	23.2	19.1	40.9	21.8	63.0
2017 Short rains						
28 days	36.1_a	57.6_a	32.1_a	37.8_a	47.6_a	49.8_a
14 days	36.2_a	57.5_a	36.0_a	45.8_a	54.0_a	56.2_a
7 days	35.1_a	51.0_a	32.9_a	36.7_a	39.5_a	45.4_a
0 days	34.7_a	72.6_a	46.2_a	48.9_a	77.1_a	56.9_a
Mean	35.5	57.5	36.8	42.3	54.6	52.1
LSD ($p \leq 0.05$)	16.5	27.5	39.1	16.5	27.5	39.1
CV (%)	20.1	19.3	34.0	20.1	19.3	34.0

Values followed by the same letter within the same row are not significantly different between the treatments using Fishers Protected LSD test ($P \leq 0.05$).

Table 3. Area under disease progress curve on KK8 and GLP2 bean varieties after incorporation of lablab green manure

Day after incorporation	2016 Short rains			2017 Short rains		
	KK8	GLP2	Mean	KK8	GLP2	Mean
28 days	211.7_b	367.8_{ab}	289.8_b	646.0_b	726.9_{ab}	686.5_b
14 days	226.7_b	387.4_{ab}	307.1_b	732.3_{ab}	831.8_{ab}	782.1_{ab}
7 days	239.4_b	433.0_{ab}	336.2_b	661.4_b	637.6_b	649.5_b
0 days	620.3_a	846.0_a	733.2_a	957.8_a	969.6_a	963.7_a
Mean	324.5	508.6	416.6	749.4	791.5	770.5
LSD ($p \leq 0.05$)	359.4	359.4	241.4	273.1	273.1	241.4
CV (%)	59.1	59.1	27.9	24.3	24.3	27.9

Values followed by the same letter within the same row are not significantly different between the treatments using Fishers Protected LSD test ($P \leq 0.05$).

3.3. Root Rot Pathogens Isolated from Bean Stems

Fusarium solani and *Fusarium oxysporum* were the most dominant root rot pathogens associated with common beans isolated from bean stem samples collected from both sites (Table 4). Other root rot pathogens isolated at both sites in both seasons albeit in small proportions were *Macrophomina phaseolina*, *Pythium ultimum*, and *Rhizoctonia solani*. The results show higher incidence of root rot pathogens in plots incorporated with lablab at planting. There was significant difference ($P \leq 0.05$) in the incidence of *Pythium*, *Fusarium oxysporum* and *Macrophomina* and between the different incorporation times across the study sites. In the first season, bean stems collected from Kapkerer had significantly higher (32%) incidence of root rot pathogens compared to stems collected from Koibem. At both sites, plots incorporated at planting had the highest population of *Fusarium*, *Macrophomina*, and *Pythium* while plots incorporated with green manure 28 days before planting had the lowest population of root rot pathogens. A significant variation in the incidence of root rot pathogen population was found between the sites according to Tukey's test at $p \leq 0.05$. In the second season, there was significant variation ($P \leq 0.05$) in the incidence of root rot pathogens isolated from different treatments at both sites. *Fusarium solani* and *Fusarium oxysporum* were the most dominant root rot pathogens isolated from stems collected from the sites, however, stems collected from Koibem had the highest contamination with root rot pathogens compared to those from Kapkerer. Other root rot pathogens isolated in small incidences include *M. phaseolina*, *Pythium ultimum* and *Rhizoctonia solani*. At both sites, bean stems collected from plots incorporated with lablab green manure at planting had the highest incidence of root rot pathogens while plots incorporated 28 days before planting had the lowest incidence of root rot pathogens. There was significant variation in the incidence of root rot pathogen population between the sites and between the treatments according to Tukey's test (ANOVA) at $p \leq 0.05$.

Table 4. Percent incidence of root rot pathogens contaminating bean stems after lablab green manure incorporation

Root rot pathogens	Weeks after incorporation									
	2016 Short rains					2017 Short rains				
	0	7	14	28	Mean	0	7	14	28	Mean
Koibem										
F. solani	60.0_a	60.0_a	66.7_a	50.0_a	59.2_a	81.1_a	64.4_a	63.3_a	58.9_a	69.9_a
F. oxysporum	48.3_a	20.0_b	23.3_a	33.3_a	31.2_b	85.6_a	67.8_a	60.0_b	61.1_a	68.6_a
M. phaseolina	45.0_a	20.0_b	25.0_a	11.7_b	25.4_b	15.6_b	12.2_b	7.8_b	15.6_b	12.8_b
P. ultimum	41.7_b	10.0_b	6.7_b	5.0_b	15.9_b	14.4_b	3.3_b	11.1_b	11.1_b	9.9_b
R. solani	25.0_c	18.3_b	0.0_b	10.0_b	13.3_b	25.6_b	12.2_a	10.0_a	13.3_b	12.5_b
Mean	44.0	25.6	24.3	22.0	29.0	44.5	31.9	30.4	32.0	34.7
LSD ($p \leq 0.05$)	15.7	24.4	32.3	23.6	22.8	27.4	20.9	26.0	28.1	39.1
Kapkerer										
F. solani	73.3_a	55.0_a	71.7_a	66.7_a	66.7_a	57.8_a	60.0_a	62.2_a	72.2_a	63.1_a
F. oxysporum	50.0_b	21.7_b	30.0_b	31.7_b	33.4_b	71.1_a	67.8_a	64.4_a	64.4_a	66.9_a
M. phaseolina	33.3_b	25.0_b	40.0_b	6.7_c	26.3_b	32.2_b	6.7_b	11.1_b	7.8_b	14.5_b
P. ultimum	13.3_c	13.3_b	18.3_b	18.3_{bc}	15.8_b	45.6_a	8.9_b	18.9_b	18.9_b	23.1_b
R. solani	41.7_b	15.0_b	25.0_b	25.0_b	26.7_b	26.7_b	8.9_b	22.2_a	5.6_b	15.9_b
Mean	42.3	26.0	37.0	29.7	33.8	42.3	30.5	35.8	33.8	36.7
LSD ($p \leq 0.05$)	15.7	24.4	32.3	23.6	24.2	27.4	20.9	26.0	28.1	32.4

Values followed by the same letter within the same row are not significantly different between the treatments using Fishers Protected LSD test ($P \leq 0.05$).

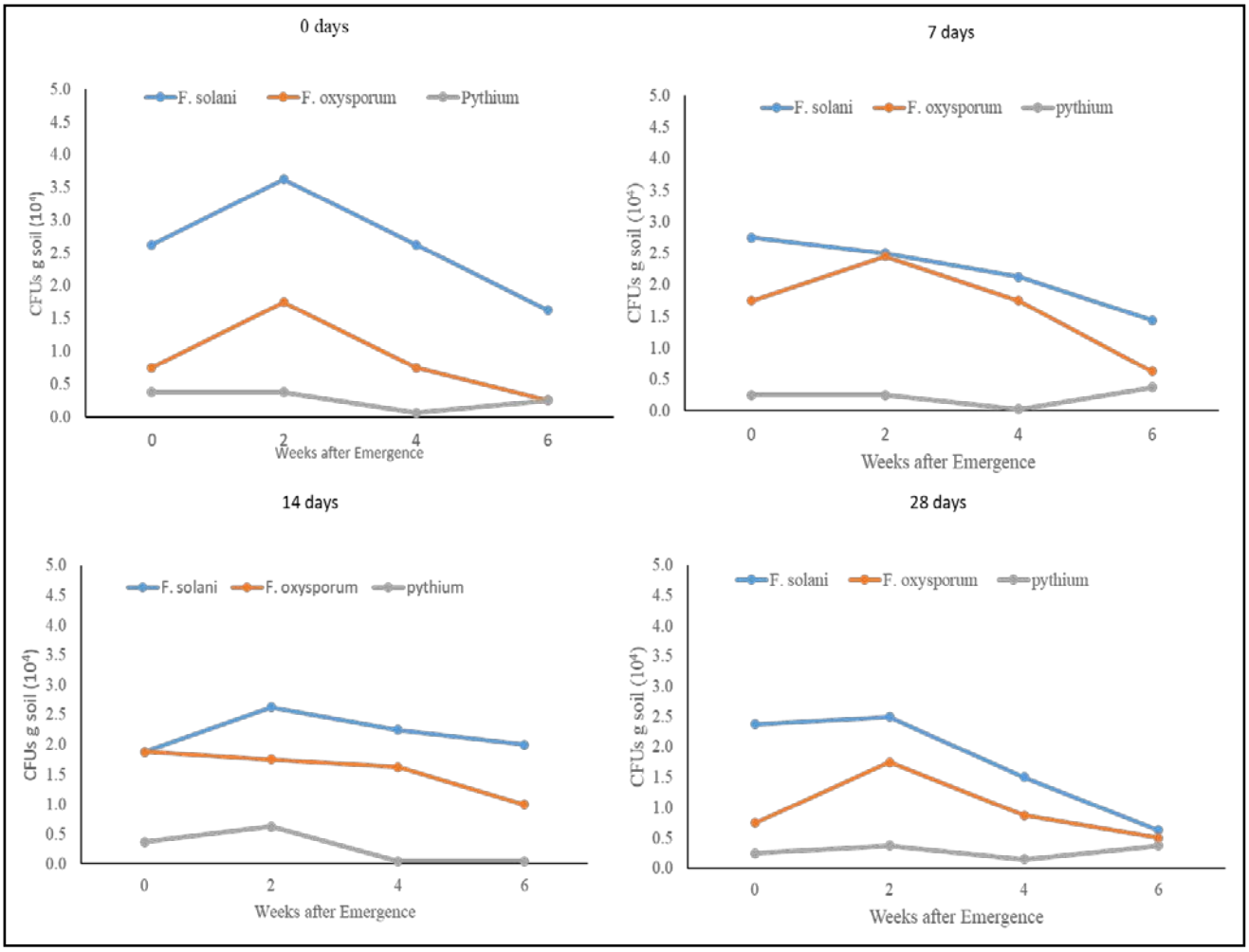

Figure 1. Population (10^4) of root rot pathogens in plots incorporated with lablab green manure at different times

3.4. Population of Root Rot Pathogens Isolated from the Soil

Root rot fungal population varied under different treatment application times (Figure 1). The highest microbial activity was observed 14 days after lablab green manure incorporation. The population of both *F. oxysporum* and *F. solani* was significantly higher in plots incorporated at planting and showed an increasing trend at the second week after crop emergence then drastically reduced four weeks later. Two weeks after emergence, in plots with lablab green manure incorporated at planting, the population of *Fusarium solani* increased from 2.5 to 3.7×10^4 cfus g^{-1}, while that of *Fusarium oxysporum* increased to 2.7×10^4 cfus g^{-1} then drastically decreased four weeks later. The population of Pythium was low but slightly increased six weeks after emergence. In plots incorporated with lablab green manure seven days before planting, *Fusarium solani* population varied from 2.8 to 1.4×10^4 cfus g^{-1}, while that of *Fusarium oxysporum* varied from 1.8 to 0.4×10^4 cfus g^{-1} soil under different sampling times. *Pythium* spp. populations in soil showed slight fluctuations, increasing between the first and the second sampling times. In plots with lablab green manure incorporated 14 days before planting, the population of *Fusarium solani* slightly increased from 1.9 to 2.6×10^4 cfus g^{-1} then reduced, while that of Fusarium oxysporum reduced from the initial population to 1.0×10^4 cfus g^{-1} soil, six weeks after crop emergence. The population of *Pythium* remained low and slightly increased from the initial population then reduced to near zero, six weeks after emergence. In plots with lablab green manure incorporated 28 days before planting, the population of *Fusarium solani* increased from 2.4 to 2.5×10^4 cfus g^{-1} soil then decreased, while that of *Fusarium oxysporum* increased to 1.8×10^4 cfus g^{-1} soil then reduced. The population of *Pythium* remained low over the entire sampling period then slightly increased six weeks after emergence.

3.5. Yield Attributes of Common Beans

Yield attributes of common beans were significantly affected by different times of lablab green manure application (Table 5). The grain yield was significantly different ($P \leq 0.05$) in both cropping seasons. In the first season, incorporation of lablab green manure 28 days before planting resulted in high grain yields with Koibem recording above 5 t/ha^{-1} while Kapkerer had paltry, at less than $1ton/ha^{-1}$. Plots incorporated at planting recorded the lowest means for biomass and grain yield at both the sites. Nonetheless, plots with lablab green manure applied seven and fourteen days after planting improved yield than plots treated with green manure at planting. At Koibem, the highest biomass was observed in plots treated with lablab green manure 14 and 7 days before planting, while at Kapkerer, plots incorporated 28 days earlier recorded the highest biomass weight. In the second season, plots incorporated 28 days earlier recorded the highest grain yield of $4.5tons/ha^{-1}$ at Kapkerer and $3.8tons/ha^{-1}$ at Koibem, while those that were incorporate at planting recorded the least. However, there were no wide variations in terms of grain yields in the second season. Plots planted immediately after incorporation gave consistently the lowest yield (average 1.0 t/ha^{-1}) across all seasons. The highest biomass was obtained from plots treated with green manure 28 days earlier at Kapkerer, while at Koibem the highest biomass was recorded in plots that had been incorporated with lablab green manure 28 days earlier. Plots planted immediately after incorporation had the lowest biomass yield at both sites in both seasons.

Table 5. The effect of different incorporation times of lablab green manure on grain and biomass yield of common bean

Days after incorporation	Koibem		Kapkerer	
	Plant Biomass (kg/ha)	Grain Yield (t/ha)	Plant Biomass (kg/ha)	Grain Yield (t/ha)
2016 Short rain				
28 Days	1176.0a	5.1a	613.0a	0.43ab
14 Days	1470.0a	5.2a	329.0a	0.48a
7 Days	1578.0a	5.1a	541.0a	0.30b
0 Days	251.0b	0.4b	181.0a	0.09c
Mean	1119.0	3.9	416.0	0.32
LSD ($p \leq 0.05$)	727.7	2.9	562.0	0.17
CV (%)	25.0	21.3	42.5	25.6
2017 Short rain				
28 Days	310.5a	3.7a	251.8a	4.3a
14 Days	236.8a	2.3ab	179.2ab	3.2ab
7 Days	123.8b	1.3ab	124.2ab	2.3ab
0 Days	116.8b	1.1b	113.0b	1.1b
Mean	196.9	3.9	167.1	2.9
LSD ($p \leq 0.05$)	102.6	3.2	102.6	3.2
CV (%)	29.7	54.2	29.7	54.2

Values followed by the same letter within the same row are not significantly different between the treatments using Fishers Protected LSD test ($P \leq 0.05$).

Table 6. Correlation coefficients between roots rot pathogens, severity of root rot, biomass and grain yield

	F. oxysporum	*F. solani*	*Pythium*	Emergence	Severity	Biomass (Kg)	Yield (ha)
F. oxysporum	-						
F. solani	0.2256	-					
Pythium	0.3149	-0.1467	-				
Emergence	-0.1532	-0.3042	-0.2551	-			
Severity	0.3239	-0.053	0.2069	-0.4505	-		
Biomass (Kg)	0.0435	-0.0481	0.0444	-0.4794	0.3514	-	
Yield (ha)	-0.1296	-0.3483	-0.2278	0.2091	0.0049	0.3468	-

3.6. Relationships between Root Rot Pathogens, Severity and Yield

For bean root rot, the isolation frequency of *F. oxysporum* and *Pythium ultimum* was positively correlated with severity ($r = 0.3239$, $r= 0.209$, $p \leq 0.05$) but was negatively correlated with emergence ($r = -0.1532$, $r= -0.3042$, $p \leq 0.05$). Seedling emergence showed negative relation with root rot pathogens and positive correlation with grain yield. However, root rot severity showed a positive insignificant correlation with yield ($r = 0.0049$, $p \leq 0.05$). However, the correlation coefficient revealed that grain yield was positively correlated with emergence ($r = 0.2016$, $p \leq 0.05$) but yield ($r = - 0.1296$, $p \leq 0.05$) showed negative and significant association with root rot pathogens (Table 6).

4. Discussion

Incorporation of lablab green manure at planting had considerable negative effects on emergence and early establishment of beans at both sites in both seasons. In contrast, plots incorporated seven days earlier had none or only slight effects on common beans emergence. Application of crop residues on top of the seed can reduce emergence, plant establishment, growth and, in some instances, yield [25,26]. The high germination percent in plots incorporated with green manure before can be attributed to high porosity, aeration, water holding capacity, and presence of humic like substances produced during decomposition [27]. The enhancement of seed germination may also be attributed to organic manure allowed to decompose ensuring availability of macro and micronutrients in soil for germination [28]. Green manure residues allowed to decompose at field capacity become harmful during the initial stages of decomposition therefore, the inhibition in germination may vary with decomposition period [29]. The effect of green manure on emergence of crops is associated with the stress experienced by seeds during green manure breakdown [30].

Delay in nutrient release by decomposing green manure, allelopathy, high osmotic potentials within decomposing residues, and increased incidences of disease following green manure crops are some of the mechanisms associated with poor crop emergence and establishment [31]. Within four to seven days of decomposition, the carbon dioxide production reaches maximum when easily decomposable green manure is incorporated [32]. Heavy application of these types of organic matter result in oxygen deficiency in the plant roots and production of substances that are harmful to plants thus reducing conditions necessary for crop growth. Composting materials before being incorporated therefore improves the physical and chemical conditions of the soil [32].

The effect of green manure on crops depend on factors such as environment, nature and quantity of the green manure, levels of moisture during decomposition and timing of sowing following green manure incorporation [31]. Green manure from cowpea incorporated before planting broccoli crop resulted in poor germination [12]. Our results confirm the need to synchronize the date of green manure biomass incorporation to the soil and crops needs. Green manure incorporation from 14 up to 28 days before planting allows for the reduction of phytotoxins from green manure thus improves crop emergence and establishment. Haramoto and Gallandt, [33] reported a reduction in crop emergence by about 23 to 33% following crimson clover green manure application and this was attributed to nitrogen immobilization or phytotoxins released. Decomposition of incorporated legume green manure may discharge large flush of ammonia that is toxic to newly germinating seedlings and at the same time slow decomposition of organic matter under drier conditions may induce inhibition of germination and initial growth of a consecutive crop [27].

High incidences accompanied with large area under disease progress curves were observed in plots incorporated at planting. The lower incidence of root rot in plots that had earlier been incorporated is due to the greater decomposability of lablab green manure. Green manure provides nutrient rich organic carbon which stimulated the population and growth of root rot pathogens. Root rot pathogens are more devastating in soils of low fertility [34] as confirmed by high isolation of root rot pathogens from Kapkerer site which is low in fertility due to intensive farming practices. From our results, low population of root rot pathogens were also observed in plots that had earlier been incorporated with lablab green manure confirming earlier findings that root rot pathogens are low when availability of soil nutrients is high [19].

Root rot pathogens isolated from stem bases of beans included *F. oxysporum*, *F. solani*, *Pythium* spp, *Macrophomina* and *Rhizoctonia* spp, high population of these pathogens were isolated in plots incorporated at planting. Continuous cropping changes soil physiochemical parameters and microorganisms thereby affecting negatively soil fertility, leading to decline in crop productivity [35]. Susceptible crops cultivated overtime increases the population density of pathogen in

soil and this is directly proportional to the disease intensity in the crop [36].

Different application times of lablab green manure had significant impacts on soil microbial community composition and structure. Among microbial populations that are reported to be related to bean health, *Fusarium* was the most abundant isolated from all the soil samples. Fourteen days after application of lablab residues, high densities of *F. oxysporum*, *F. Solani* and *Pythium* spp. were observed. Gomez *et al*., [37] reported similar fungal species in their work and they established that the pathogens predominated over other fungal populations because they are capable of using different substrates in the soil. The populations of microorganisms in soil are influenced by nature of green manure, degree of decomposition, carbon: nitrogen ratio, time of application and amount of fresh organic residues applied [38]. The quality of the organic matter determines whether beneficial or facultative pathogens multiply fast [39]. Fresh or barely decomposed organic residues may lead to short-term upsurge in populations of soilborne pathogens such as *Rhizoctonia* and *Pythium* spp. because they reproduce easily in such materials. Van Bniggen and Termorshuizen [40]. reported an increase in severity of Pythium damping-off between 7 – 10 days after incorporation of cover crops. In view of the risks associated with green manure incorporation, although temporary, the timing of incorporation in relation to planting is very critical.

There is much difference between fresh green manure and those that are partially decomposed. Increased disease pressure following green manure application is because organic manures provide environments that are not conducive for growth and explosion of antagonistic microorganisms which is characterized by high salt and ammonium concentrations, and lack of oxygen [39]. Use of composts that vary in salinity, nitrogen availability, and degree of decomposition may lead to increase in disease incidence and severity [40]. Direct incorporation of lablab amendments increased fungal population initially, indicating that the fresh lablab promotes long-term stimulation of the microbial population. According to Elfstrand *et al*., [41] the increase in population could be due to high availability of carbon compared with the processed forms fertilizer, resulting in faster growth of microorganisms after direct incorporation. They further noted that, timing of incorporation is the cause for the observed differences, since direct incorporation of the red clover ley was carried out two weeks before the incorporation of slurry and compost.

The correlation analyses generated significant results between different parameters. Thus root rot pathogens positively correlated with severity but were negatively correlated with emergence, while seedling emergence showed negative correlation with root rot pathogens and positive correlation with grain yield. This indicates that high population of root rot pathogens would necessarily induce pre and post emergence damping off. Positive effect of green manure was found in plots treated with lablab green manure before planting. Yield results from the present study showed that adequate decomposition time allows for adequate release of nutrients for uptake by plants. This confirms findings by Talgre *et al*., [42] who reported that the effect of green manure residue become apparent later. Biomass from green manure legumes adds considerable amount of nitrogen to the soil but it is released overtime and this ensures that the positive effect of green manure may be felt for a long time [42]. The results of the study indicated a negative correlation between root rot pathogens, severity and grain yield exist so it can be predicted that with an increase in root rot pathogen population, root rot severity and incidence increases and this in turn results in yield loss.

5. Conclusion

The efficiency of green manure depends on the time of biomass application therefore residues must be allowed to decompose sufficiently and colonized to a degree that allows microbiostasis since immature residues contain toxic compounds that affect growth of plants and predispose plant roots to attack by diseases.

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Statement of Competing Interests

The authors declare no competing interests.

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