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Full Length Research Paper

Impact of crop rotation sequences on potato in fields inoculated with bacterial wilt caused by *Ralstonia solanacearum*

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The potato industry in Kenya is threatened by bacterial wilt because most production areas are infested with the wilt-causing *Ralstonia solanacearum* and over 50% yield losses have been reported. Continuous cultivation causes soil physical and biological constrains that greatly affect the crop performance and increase proliferation of the bacterium. Rotation with non-host or suppressant plant species could contribute to considerable reduction of bacterial wilt in the subsequent potato crops. This study tested the effect of different crop sequences on *R. solanacearum* population in the soil, wilting incidence and yield of potato. Two season field experiments were conducted at two sites (Egerton University, Njoro and National Agricultural Research Laboratories (NARL), Kabete) with 17 different crop sequences. Rotations involving brassica and legumes with potato gave a higher emergence percentage compared to the other sequences. The bacterial population was significantly influenced by the different environments from the first season to the third season; F (1, 102) =53.2, P<0.001, F (1, 102) =12.5, P< 0.001 and F (1, 102) =236.8, P<0.001 respectively. There was a significant effect F(16,119)=7.063, P<0.001 of the crop rotation sequences on the wilting incidence of potato. Pre cropping potato with spring onion and barley resulted to a significantly lower wilting incidence compared to all the other crop rotation sequences with a mean of 8.3% across sites. The results showed that Potato-Lablab-Potato and Cabbage-Lablab had the highest yield with 19.9 and of 19.7 tons/ha in the one crop rotations and pre crops to potato respectively. A Genotype x Environmental means versus IPCA scores showed that the yield due to barley-spring onion, spring onion-barley and wheat-spring onion as pre crops were more stable in both locations compared to the other cropping sequences. The study indicates that rotations involving spring onion with the locally grown cereals such as barley and wheat can be utilized in curbing bacterial wilt. Rotations involving lablab and cabbage may also be used to increase the yield of potato in bacterial wilt infested fields. These crops should be used in rotations involving more seasons so as to achieve better effects.

Key words: Bacterial wilt, crop rotation, potato yield, *Ralstonia solanacearum*, wilting incidence.

INTRODUCTION

Potato crop is the second most important staple food crop after maize in Kenya and an important food and cash crop in the medium and high rainfall areas where it has a comparative advantage over maize. Continuous cultivation on agricultural land causes soil physical constraints in the form of high bulk density, low water conductivity and reduced effective rooting depth of crops, which can greatly affect crop performance and increase nematodes infestation (Akanni and Ojeniyi, 2008). Bacterial wilt caused by *Ralstonia solanacearum* is reported to be one of the major challenges affecting potato farmers in Kenya (Ateka et al., 2001; Kaguongo et al., 2009; Nyangeri, 2011; Kwambai et al., 2011). Small scale farmers own small land parcels that limit them from practicing crop rotation and therefore practice continuous cultivation of crops in bacterial wilt infested soils. The question is therefore; what kind of cropping systems can be adopted by small scale farmers to reduce bacterial wilt infection in potato crop and thereby strengthen potato production?

Crop rotation as a cultural control method is applied with the objective of achieving maximum benefits due to the contribution of the crops to the soil and the crops effect to the pathogen population. The use of these crops is based on several principles such as use of non host crops, crops with suppressive effect due to their exudates and secondary metabolites, N fixing plants, plants with high residual matter and their adaptability to the specific environs. Plants release secondary metabolites in significant amounts at varying stages of plant growth. Some of the reported metabolites include; Benzoxazinoids: 2,4-Dihydroxy-7-methoxy-2H-1,4-benzoxazin-3(4H)-one (DIMBOA) which is the most common benzoxazinoid found in wheat and wild barley among other cereals (Fall and Solomom, 2011). It is reported to cause allelopathy, repel insects and also resist pathogens. They are stored as inactive glucosides in the plant but become active upon tissue disruption. They cause mutagenic effects on the pathogens DNA and react with amino acids. Flavonoids are present in barley in the form of lutonarin, saponarin and isovitexin. Several phytoanticipans in the flavonoids group are reported to inhibit spore germination and the growth of *Xanthomonas oryzae* (Padmavati et al., 1997). UV-absorbing flavonoids in cotton leaf tissues have also been found to be antagonistic to *Xanthomonas campestris* (Edward et al., 2008).

Host exclusion or withdrawal is achieved by growing non hosts crops in any crop rotation regimes. The duration taken in the crop rotations highly determines the reduction of inoculum in the soil. Short term crop rotations have been shown to affect pathogen populations and disease status in the soil (Lemaga, 2001; Narayanasany, 2013). However research on long term rotations have shown that crop rotations require more time to significantly and effectively reduce soil borne pathogens (Larkin et al., 2010; Wright et al., 2015). Rotational crops are also used as cover crops to suppress disease both in the growing phase and in the decomposition phase by inducing an increase in soil microbial biomass and in soil biological activity. The nutritive contribution of the rotation crops as cover crops and as green manure is also another factor that is utilized in crop rotations. Residue quality is known to indirectly influence the organic matter content and aggregation. A marked difference has been observed in the levels of carbon in the soil after root and shoots were incorporated into the soil (Ball et al*.,* 2005). Shoots are considered to breakdown rapidly compared to the roots and therefore are a short term source of nitrogen to the subsequent crop. Decomposition of incorporated crop residues also vary depending on the C:N ratio and lignin content of the crop residues. Residues with a low C:N ratio (<25:1) are shown to decompose rapidly thereby creating a suitable substrate for microbial activity (Kriaučiūnienė et al., 2012). Dolichos bean has been found to decompose rapidly with a reduction of at least 25% to 63% of their initial dry weight within the first four weeks (Ibewiro et al., 2000; Ruiz-Vega et al., 2010) compared to the other crops in the study.

Other dynamics that influence the disease status in the soil as a result of crops grown include: Chemotactic effect of exudates, organic and amino acids from the different cover crops or their green manure (Yao and Allen, 2006). Chemotaxis as a factor that promotes the proliferation of the pathogen and contributes to the infection rate of the host plant in a crop rotation system. It is reported that *R. solanacearum* strain K60 was attracted both to plant root exudates of tomato which is a host plant and rice , a non-host plant to the pathogen, however tomato root exudates indicated three times stronger attraction compared to rice exudates at protein concentrations of 100 ug/ml (Yao and Allen, 2006). In this study, non-host acids and root exudates are reported to be less attractive or repellant to *R. solanacearum*. Proliferation of different microbes is also influenced by the different cover crops or green manure and has been observed to vary significantly according to the type of cover crop (Patkowska and Konopiński, 2014). Other factors such as the formation of DNA-containing extracellular traps by

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Table 1. Climatic and edaphic characteristics of the experimental sites.

Site	Altitude, Longitude and Latitude	Rainfall (mm)	Temp (°C)	Soil type	pH
Egerton	2225 m asl, 01° 13'S and 35° 30'E	1012	22	Sandy loam Mollic phaozems	5.5-6.0
NARL	1737m asl, 36° 41'E and 01° 15'S	980	23	Clay loam Humic nitisol	4.5-7.0

Source: Jaetzold and Schmidt (1993) and Oloo et al. (2011).

non host plants as observed by Tran et al. (2016) and Hawes et al. (2016) also plays a role in the proliferation of disease in the soil.

This study focused on the impact of crop rotation sequences of several cereals, legumes as well as spring onion on reducing bacterial wilt in infected soils and to improve potato yield. The efficacy of any crop rotation is determined by the type of soil, soil pH, soil moisture, weather and other abiotic factors and therefore crop yield and disease incidence may vary from one location to another and from one season to another (Adebayo and Ekpo, 2006). It is therefore important to consider crops for rotation in any specific environment based on their adaptability. The present study therefore attempted to use the mentioned crops grown by small scale farmers in medium to high altitudes areas to evaluate their impact on potato production and bacterial disease when utilized in crop rotations. This was carried out in fields inoculated with *R. solanacearum* to simulate farmers fields infested with bacterial wilt. The objective of this study was to identify alternative crops besides the potato monoculture that can be used to reduce the effect of bacterial wilt and consequently increased yields.

MATERIALS AND METHODS

Site description

Two season field experiments were conducted at two sites: Egerton University, Njoro and National Agricultural Research Laboratories (NARL), Kabete field stations in the years 2013 and 2014. The edaphic characteristics of these locations are indicated in Table 1.

Preparation of inoculum

For purposes of inoculum preparation, the procedures of Kinyua and Miller (2012) were followed. *R. solanacearum* isolated from infected tubers was grown on Tetrazolium Chloride (TZC) agar medium at 28°C for 48 h. Wild-type bacterial colonies (based on colony morphology) were harvested, suspended in CPG liquid culture (Casamino acids-peptone-glucose) and incubated for three days at room temperature, Kinyua and Miller (2012). The cultures were centrifuged at 10,000 rpm for 10 min at 10°C, suspended in distilled water and adjusted to 10^8 CFU ml^{-1}, as described by Yadessa et al. (2010).

Inoculation of experimental fields

The experimental fields were inoculated with *R. solanacearum* inoculum to simulate the farmers' fields which are infested with the pathogen. Disease infested plots were developed by growing a susceptible potato variety "Tigoni" for a season. Ten mililitres of the inoculum was sprayed to the rhizosphere of each plant at the 30^{th} day after planting (Broekhuizen, 2002; Ayana et al*.,* 2011). All the potato plants were ploughed and incorporated into the soil after more than 50% of the plants showed wilting symptoms.

Treatments

The experiment had 17 crop sequences as the treatments using short season crops. Crops used were; spring onion var. Green bunching, Garden pea var. peas plum, Potato var. Tigoni, Wheat var. Kwale, Barley var, Sabini, Canola var. Tower, Lablab (Local variety) and cabbage var. Coppenhagen. The treatments were Potato-Cabbage-Potato (1), Lablab-Cabbage-Potato (2), Potato-Canola-Potato (3), Garden Pea-Canola-Potato (4), Potato-Lablab-Potato (5), Cabbage-Lablab-Potato (6), Potato-Garden Pea-Potato (7), Canola-Garden Pea-Potato (8), Potato-Spring Onion-Potato (9), Wheat-Spring Onion-Potato (10), Potato-Wheat-Potato (11), Spring Onion-Wheat-Potato (12), Potato-Barley-Potato (13), Barley-Spring Onion-Potato (14), Spring Onion-Barley-Potato (15), Potato-Potato-Potato (16) also referred to as monoculture in the text, Fallow-Fallow-Potato (17) – the plots were not ploughed or weeded in this treatment throughout the fallow seasons.

Experimental layout and agronomic practices

The experiment was laid out in a randomized complete block design with four replicates in plots of 3 m by 3 m. The two cropping patterns were considered. Rotation of potato with one crop termed as one crop rotation and use of two different crops before the main potato crop considered as pre crops to potato. The crops were planted in the long rains of 2013, short rains of 2013, and long rains 2014 in succession as indicated in the above treatment sequences. After every harvest, the above ground vegetative biomass was left on the ground and incorporated in the soil during the planting of the next crop in the next season using a hand hoe. The seed potato ("Var. "Tigoni") was planted at a spacing of 75 cm by 30 cm giving a total of 40 plants per plot in furrows in the last season. Hand weeding was done at 4 weeks after planting and ridging (earthing up) was done twice at four and eight weeks after planting. Late blight was controlled with Dithane-M45 and Ridomil-MZ 72 sprayed at alternating times at a rate of 50 g/10 L of water when it was necessary.

Data collection

Wilting incidence

Assessment of the bacterial wilt incidence started at the onset of wilt symptoms after which counting of wilted plants was done on a weekly basis. Plants that showed either complete or partial wilting were considered wilted and tagged to avoid double counting in subsequent assessments and also to avoid the possibility of missing out those completely killed early in the growth period. Wilt incidence for each treatment was calculated as number of wilted plants expressed as a percentage of the total number of plants emerged.

Bacterial population in the soil

The population of *R. solanacearum* in the soil was established three weeks after planting in the first season and at 15 weeks after planting of each crop in the subsequent seasons. Four soil samples were randomly picked from each experimental plot at 20 cm soil depth and were mixed thoroughly to make one sample. Ten grams of soil from each sample was put in a flask with 30 mls of distilled water. The soil suspension was stirred on a rotary shaker at 150 rpm for 30 min. The soil suspension was allowed to settle and 1ml aliquot suspension was drawn out using a sterile pipette tip. This suspension was put in sterile Eppendorf tubes and formed the stock suspension. Serial dilution was carried out upto 10^{-4} suspension. An aliquot of 100 µl (0.1ml) of the soil suspension was lawn plated on Semi Selective Medium (SMSA) in a petri plate for 10^{-1} and 10^{-3} serial dilutions suspensions. The plates were incubated at 30°C for 48-72 h. The colonies that showed typical *R. solanacearum* characteristics (fluidal and irregular with a characteristic red or pinkish red centres and whitish periphery) were counted from the 10^{-3} serial dilution suspension. Data of bacterial population was considered after the second crop to evaluate the impact of the two crops before the main potato crop. The number of colonies per ml was calculated using the following formula

$$No.of\ bacteria/ml = \frac{CFU \times 1000}{0.1}$$

Log transformation of bacterial population data after the second crop in the rotations was done for the purpose of analysis of variance (Log10) cfu*10000).

Potato yield

Harvesting was done once at 110 days after planting. Total weight of all tubers; ware (>55 mm), seed (35-55 mm), and chatt size (<35 mm) in diameter was recorded for each plot.

Data analysis

Data was analyzed for the response variables (Emergence %, Wilting Incidence Days After Planting (WI DAP %) and yield (tons/ha) using two way analysis of Variance (ANOVA). Post hoc mean separation was done using Tukeys HSD whenever there were significant results. Spearman *rho's* correlation was done to examine the relationships between the response variables. IBM SPSS statistic software Version 20 was used for the analysis of this data. To determine the stability of the crop sequences in the two environments, AMMI analysis was done and an IPCA versus Genotype x Environmental means plot was generated to graphically visualize the mean performances and stability of the cropping sequences on the yield of potato (GENSTAT Version 15).

RESULTS AND DISCUSSION

Effect of the crop sequences on the emergence percentage of the potato

The crop sequences did not indicate any significant effect in the emergence of the potato crop grown in the third season of the crop rotation sequences. Rotations involving brassica and legumes gave higher emergence percentage compared to the other treatments in both rotation sequences and in both sites in the subsequent potato crop (Table 2). Cabbage-Lablab and Garden pea-Canola as pre crops recorded the highest emergence percentage in the subsequent potato crop. This may be attributed to the rapid decomposition and mineralization of vegetables and the contribution of *Dolichos lablab* to fertility in the soil as also observed by Aganga and Tshwenyaye (2003), Sanginga (2003) and Agneessens et al. (2014).

Rotations involving wheat and spring onion generally gave a lower emergence percentage compared to the brassica-legume rotations. This may be attributed to the allelopathic effect reported in wheat. It contains alleochemicals such as phenolics and alkaloids found in the leaves, roots, seeds and roots which have been shown to have suppressive effects in the germination of several crop seedlings. *In-vivo* and *in-vitro* trials have shown the efficacy of cereals such as barley in suppression of germination in most seedlings such as lettuce, bread wheat, cabbage and alfalfa (Kremer and Ben-Hammouda, 2009). Wheat straw has been known to have a positive allelopathic effect in the reduction of the density and biomass of weeds. It inhibits the growth and yields of other crops such as rice, barley, cotton and soybean (Lam et al., 2012). A secondary metabolite, benzoxazinoids found in wheat undergoes enzymatic and chemical degradation upon tissue disruption and their phytotoxic mechanism is attributed to mutagenic effects on DNA and their ability to react with amino acids and disrupt proteins of germinating seedlings (Fall and Solomon, 2011). These factors may have contributed to the differences observed in the germination of potato in the third season in the different crop sequences.

Bacterial density in the soil

The bacterial population was significantly influenced by the location from the first season to the third season; F (1, 102) =53.2, P<0.001, F (1, 102) =12.5, P< 0.001 and F (1, 102) =236.8, P<0.001 respectively which is attributed to the different environmental (rainfall, temperature and soil) parameters. A major factor that contributed to the significant effect of the location as a main effect was the soil pH (Table 1). The strongly acidic clay soils are reported to favour the survival of *R. solanacearum* (Sharma, 2004) and this contributed significantly to the high wilting index in NARL site.

There was a decreasing trend of the mean bacterial population from the first season to the third season in both crop rotation patterns (Figure 1). The mean wilting incidence (WI) and average bacterial population of *R. solanacearum* was high in crop sequences involving only one crop in rotation with potato as compared to the

Table 2. Effect of different crop rotation sequences on emergence, yield and WI (%) of potato in third season

Treatments	Emergence (%)		Yield (Tons/ha)		WI 5 WAP		WI 10 WAP		WI 15 WAP	
	Eger	NARL	Eger	NARL	Eger	NARL	Eger	NARL	Eger	NARL
Spring onion-barley-potato	86.3±8.2	63.1±16.7	29.6±4.5^{ab}	7.8±2.0^{a}	0.0±0.0^{a}	0.0±0.0	0.7±1.3^{a}	0.8±1.7^{a}	2.2±2.9^{a}	14.2±4.8^{a}
Spring onion-wheat-potato	80.6±11.3	65±6.1	27.5±5.1^{ab}	7.8±1.6^{a}	2.1±2.7^{a}	1.0±0.0	4.0±4.6^{a}	1.1±2.2^{a}	6.1±6.0^{ab}	14.1±8.6^{a}
Canola-garden pea-potato	79.4±12.5	65.6±10.8	30.9±2.6^{a}	7.1±0.25^{ab}	1.5±3.0^{a}	0.0±0.0	3.6±4.2^{a}	0.8±1.7^{a}	5.3±2.2^{ab}	15.0±11.5^{a}
Barley-spring onion-potato	80.6±6.9	72.5±10.2	28.9±2.3^{ab}	7.5±0.9a^{b}	0.7±1.5^{a}	0.0±0.0	3.0±2.4^{a}	0.8±2.5^{a}	6.1±4.1^{ab}	16.7±3.6^{a}
Lablab-cabbage-potato	84.4±4.5	70.6±8.7	31.4±3.1^{a}	6.5±1.4^{ab}	6.0±6.9^{a}	0.0±0.0	6.5±10.5^{a}	0.8±2.2^{a}	11.2±10.1^{ab}	13.4±3.7^{a}
Potato-barley-potato	84.4±5.5	61.8±10.1	32.0±4.3^{a}	4.3±2.4^{ab}	2.9±4.1^{a}	0.0±0.0	5.6±4.7^{a}	0.0±0.0^{a}	6.6±3.5^{ab}	18.5±8.1^{a}
Potato-lablab-Potato	83.3±7.5	69.4±14.2	33.2±3.6^{a}	6.7±2.3^{ab}	3.6±4.5^{a}	0.0±1.6	5.7±7.7^{a}	0.8±1.6^{a}	8.7±7.3^{ab}	17.1±12.0^{a}
Potato-wheat-potato	83.3±9.2	61.2±10.9	25.8±4.7^{ab}	5.4±3.8^{ab}	6.6±4.7^{ab}	0.0±0.0	10.8±9.1^{a}	4.3±6.3^{ab}	13.9±5.8^{ab}	12.8±5.4^{a}
Cabbage-lablab-potato	85.6±4.3	72.5±8.9	33.0±5.6^{a}	6.5±0.5^{ab}	9.7±4.3^{ab}	0.0±0.0	11.7±3.9^{a}	0.8±1.6^{a}	13.1±4.7^{ab}	17.1±12.0^{a}
Potato-garden pea-potato	80.0±16.2	60.6±8.8	28.7±2.7^{ab}	6.8±1.1^{ab}	4.7±2.8^{a}	0.9±1.8	4.7±2.8^{a}	0.9±1.8^{a}	12.0±7.0^{ab}	17.6±11.6^{a}
Potato-spring onion-potato	77.5±11.9	63.8±13.6	29.4±2.3^{ab}	4.8±3.2^{ab}	3.1±2.5^{ab}	0.0±0.0	5.6±2.8^{a}	1.9±2.3^{a}	8.3±4.2^{ab}	21.9±11.8^{a}
Wheat-spring onion-potato	71.9±4.3	69.4±9.4	29.2±2.9^{ab}	6.7±1.5^{ab}	6.4±6.1^{ab}	0.0±0.0	9.6±7.3^{a}	2.0±4.0^{a}	16.5±13.1^{ab}	15.0±6.0^{a}
Fallow-fallow-potato	70.6±12.3	73.1±14.8	28.5±4.2^{ab}	8.3±2.2^{a}	8.3±9.0^{ab}	0.8±1.5	8.3±9.0^{a}	0.8±1.5^{a}	15.1±9.9^{ab}	16.9±2.2^{a}
Garden pea-canola-potato	79.4±8.2	80.0±9.1	26.7±2..4^{ab}	8.9±1.2^{a}	6.9±5.4^{ab}	0.0±0.0	10.0±8.1^{a}	1.4±1.6^{a}	10.6±7.9^{ab}	15.9±5.3^{a}
Potato-canola-potato	80.6±7.7	78.8±4.8	28.7±3.2^{ab}	7.8±1.2^{a}	6.0±7.1^{a}	0.0±0.1	11.4±9.2^{a}	2.4±1.6^{a}	13.8±9.2^{ab}	24.3±9.3^{a}
Potato-cabbage-potato	78.1±8.3	77.5±6.1	26.4±6.3^{ab}	7.6±1.2^{ab}	11.8±3.3^{ab}	0.8±1.6	19.5±7.9^{ab}	2.4±2.9^{a}	23.4±10.4^{bc}	24.5±12.2^{ab}
Potato-potato-potato	79.4±8.3	70.6±12.9	20.7±3.1^{c}	2.7±2.3^{b}	22.7±15.8	0.8±1.6	36.5±9.9^{b}	10.5±4.8^{b}	39.4±10.4^{c}	45.5±2.4^{b}
Mean	**80.4**	**69.2**	**28.8**	**6.6**	**6**	**0.3**	**9.9**	**1.9**	**12.5**	**18.7**
Pvalues	**0.568**	**0.242**	**0.01**	**<0.01**	**<0.01**	**0.19**	**<0.001**	**<0.001**	**<0.001**	**<0.001**

results from this study also confirm that cabbage is not a recommendable crop to use for rotations in bacterial wilt infected fields. The study also indicates that barley-spring onion has potential to suppress diseases and concurs with similar results from a previous study showing barley/clover causing a reduction of the fungal diseases in the short term (Larkin et al., 2010). The effect of barley and wheat in the suppression of disease in the subsequent crop and reduction of the bacteria population may be attributed to root exudates microbial interaction. The study also highlights a scientific question whether the order of the crops has an interactive effect on disease or pathogen inoculum. According to the study pre crops starting with spring onion resulted to a lower wilting incidence compared to the vice-versa.

Previous studies have shown a positive contribution of brassicas such as canola to reduction of fungal soil borne diseases (Bohinc et al., 2012; Boydston et al., 2011; Larkin et al., 2010; Bednarek et al., 2009). However, incorporation of canola as green manure in this study did not have a significant effect on bacterial wilt nor on the *R. solanacearum* population in the soil.

The bacteria population after the second season negatively correlated (spearman's rho) with the emerged plant stand of the potato crop in the third season (*rs*=-0.135). The emerged plant stand significantly correlated with the yield at (r_s= 0.565, p<0.001) and an increase in the wilting incidence resulted to a significant decrease in the yield (r_s=-0.380, p<0.001). This indicates the significance of the bacterial population on the potato crop from emergence to harvesting.

Effect of the crop rotation sequences on the yield of potato planted in the third season

There was a significant interaction between the

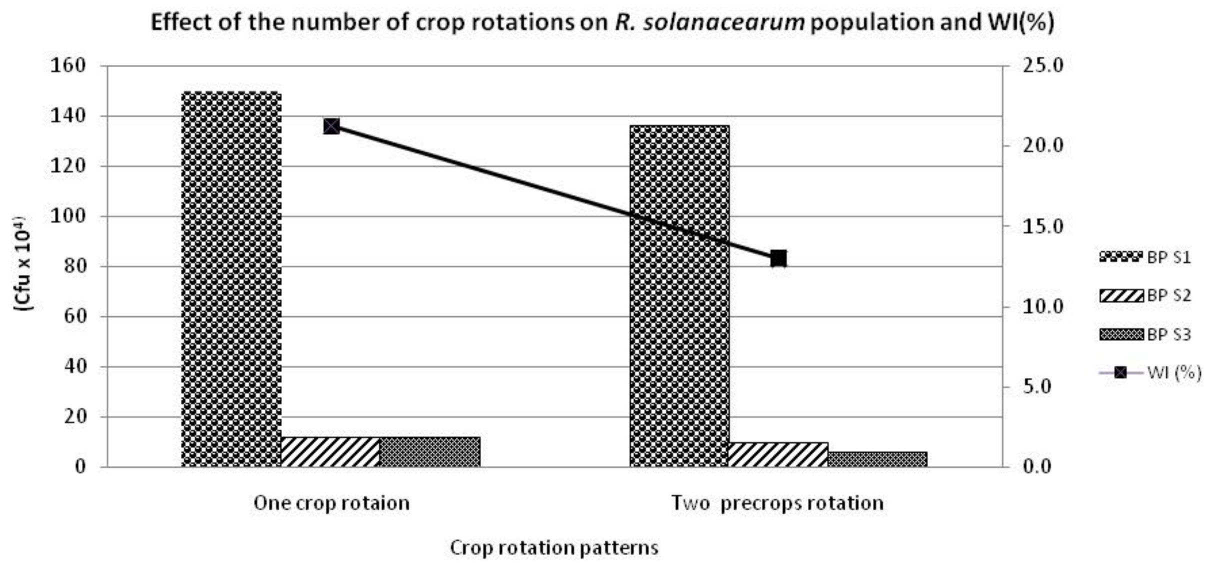

Figure 1. Effect of one crop rotation and pre crops sequences to potato on *R. solanacearum* population and WI% in the main potato season. Values are means of the cropping sequences in each cropping pattern. BP: Bacteria Population; S: Season; WI: Wilting incidence.

sequences involving two pre crops to potato. The mean yield was also higher in the two pre crops rotations compared to the one crop rotations with potato. The lower wilt incidence due to two seasons of rotation with different crops (Precrops) may be attributed to the break period which is a physical mechanism in controlling pathogen effect by withdrawal of the host. The absence of the host for two seasons in the two pre crop rotations resulted to a higher decline in the pathogen density compared to the one crop rotation sequences.

The bacterial population after the second crop was considered so as to evaluate the impact of the rotation crops on bacterial population in reference to the initial inoculation stage (Figure 2a and b). The pre crop of wheat-spring resulted to the highest decline of the *R. solanacearum* density in the soil (Figure 2b). A combination of the allelopathic effect of wheat and onion may have contributed to the reduced density of *R. solanacearum* in comparison to the other crop sequences. Flavonoids and benzoxazinoids are found in cereals such as wheat and barley and they have been reported to inhibit spore germination and growth of several pathogens such as *Xanthomonas oryzae* and *Xanthomonas campestris*, (Padmavati et al., 1997; Edward et al., 2008; Fall and Solomom, 2011).

Wilt incidence in the potato crop

The wilting progress significantly varied with the crop sequences. The progression of wilt in potato-cabbage-potato rotations did not significantly differ from the potato monoculture-no rotation (negative control) across all the three seasons as observed in Table 2. All the other one crop rotations had significantly lower progress in the disease development compared to the potato-cabbage-potato and the monoculture. There was a significant main effect of the crop rotation sequences on the wilting incidence; F (16,119) =7.063, P<0.001. Pre cropping potato with spring onion and barley resulted to a significantly lower wilting incidence compared to all the other crop rotation sequences with a mean of 8.3 and 2.2% wilt incidence respectively in both sites. Rotation of potato with wheat alone also had the least wilting incidence (12%) at NARL site. Spring onion-wheat-potato also had a lower wilting incidence of 10.1%, mean in both sites. Among the *R. solanacearum* non host bulb type crops recommended for crop rotation include spring onion according to Wang and Lin (2005). Potato-barley-potato rotation had the least WI in the one crop rotations with a mean of 12.1% across the two sites. Rotation of potato with cereals such as wheat and barley and spring onion was found to consistently influence the reduction of bacteria population and consequently reduce the wilting incidence (Table 2). Data in the table shows that the wilting incidence progress accelerated from the 10th week onwards.

Cabbage (*Brassica oleraceae* L.) is reported to be a common host plant of bacteria wilt and has also been found to be infected by *R. solanacearum* biovar 2 and 3 (Alvarez et al., 2008; Guidot et al., 2014; Nortj'e, 2015). Evaluation of different plants on wilt incidence and infection by *R. solanacearum* biovar 2 and 3 showed *Brassica oleraceae* var. capitata is a host to the bacterium whereas spring onion did not show any wilting nor was it infected by the two biovars (Nortj'e, 2015). The

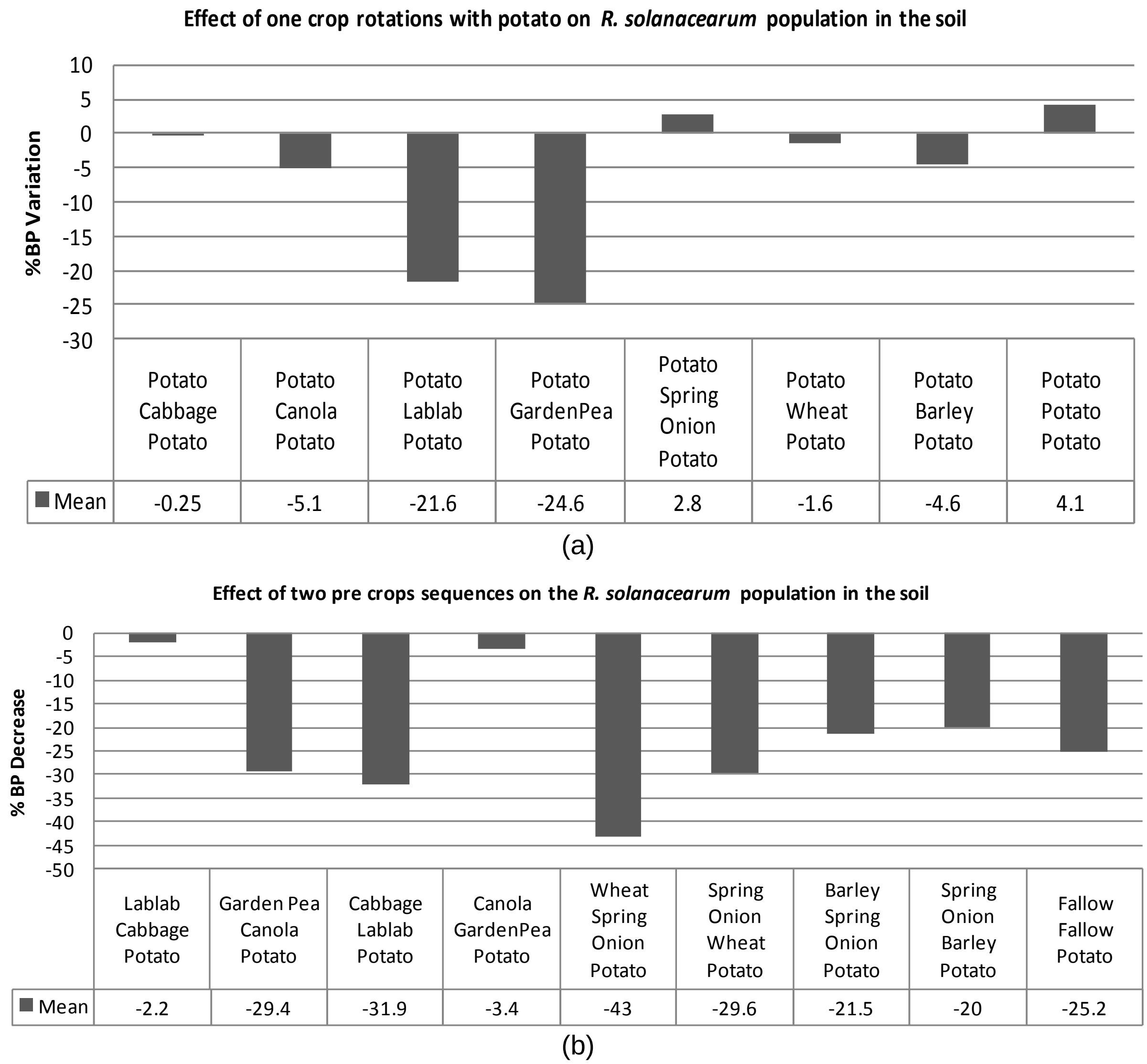

Figure 2. a) Effect of the different pre crops sequences on the % bacteria population in the soil after the second crop/season. b) Effect of the Pre crop rotations on the % bacteria population in the soil after the second crop % BP:% Bacterial Population % (Negative or positive figures denote a decline or an increase in the *R. solanacearum* population density respectively in reference to the bacterial population density after inoculation of the experimental field before the first crop was planted).

cropping sequences and the location $F(1,102)=1.9$, $P<0.025$ on the yields. The yield was significantly lower in NARL compared to Egerton site. This is attributed to the acidic soils (Table 1) found in NARL which favour proliferation of *R. solanacearum*. The mean yield in both sites showed that all other crop rotation sequences were significantly different from the monoculture except for the one crop rotation of potato with cabbage, wheat and spring onion. Potato-lablab potato and potato-canola-potato had the highest yield with 19.9 and 18.2 tons/ha respectively in the one crop rotations across the two sites. Cabbage-Lablab and Lablab-Cabbage as pre crops to potato also resulted to the highest yield of 19.7 and 19.0 tons/ha across the two sites. As shown in Table 2, rotation of potato with Lablab and canola yielded the highest in Egerton and NARL respectively in the one crop rotations with potato. Lablab is able to transport minerals from the depths of the soil to make it available to the plants due to its deep tap root and its active role in N fixation in the soil due to the presence of N fixing bacteria (Aganga and Tshwenyaye, 2003; Sanginga, 2003). Legumes are known to form symbiotic relationships with soilborne rhizobia known as plant growth promoting rhizobacteria (PGPR). These plant growth promoting bacteria produce plant growth regulators, are involved in symbiotic N fixation and solubilize minerals such as

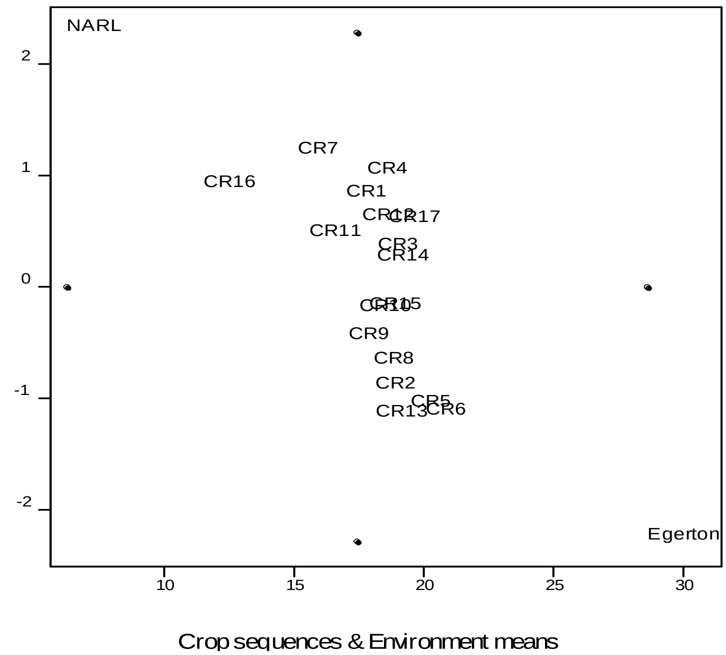

Figure 3. AMMI model for potato yield (t/ha) showing the genotype and environment means (X axis) against their respective IPCA 1 scores (Y axis).

phosphorus among other beneficial mechanisms that are important to crops (Montano et al., 2014; Cooper, 2008).

The presence of vegetable in the brassica legume patterns in the best yielding crop sequences may also have played a role to the yield increase due to the short duration taken by most vegetables for mineralization after incorporation into the soil surface (Agneessens et al., 2014). A study by Nyangeri (2011) indicated that cabbage recorded significantly marketable yields compared to crops such as maize and beans in one crop rotations with potato. Another research evaluating the rotation effects of canola, barley, and green beans to potato yield showed that canola yielded significantly higher yields compared to the other crops (Larkin et al., 2010). This study concurs with these previous results indicating the potential of brassicas in rotation with potato in increasing tuber yield. Residues with a high C:N ratio, high lignin and polyphenols content are known to immobilize inorganic N, resulting to reduced microbial activity and therefore reduced yields (Kumar and Gor, 1999). This may have contributed generally to the lower yields in the wheat and barley rotations with potato when compared to the rotations with legumes and brassicas.

Stability of the crop sequences

An additive main effects and multiplicative interaction (AMMI) model which combined the analysis of variance for the crop sequences and location main effects with the principal component analysis (PCA) of the crop sequences-environment interaction was performed on yield data of the potato (Figure 3). The effectiveness of crop rotation is highly dependent on the prevailing weather characteristics of any environment, adaptability of crops being used in the rotations and the soil environments which comprises of the physical and biochemical properties of soil. The potato yield after barley-spring onion (CR14), spring onion-barley (CR15) and wheat-spring onion (CR10) as pre crops was more stable in both locations compared to other cropping sequences. Rotation of potato with garden pea was more stable in NARL while cabbage-lablab and rotation of potato with lablab and barley were more stable in Egerton. The concept of environment specific system has also been demonstrated by several authors (Seremesic et al., 2013; Mrabet, 2011).

Conclusions

Rotations involving brassicas and legumes gave higher germination in the study. A higher emergence translated to a higher yield. Canola can be used in rotations with legumes with an objective of increasing the emergence percentage of potato. Two pre crops to potato will result to a decrease in bacteria population and at the same time reduce wilt compared to one crop rotation. The study also indicated that two successive crops planted as pre crops to potato are not adequate to destroy all the *R. solanacearum* population or eradicate the disease in the soil especially so if the inoculum is high. Rotation involving spring onion and cereals such as barley and spring onion can be recommended to farmers in reduction of the bacteria population and wilting incidence in the subsequent potato crop but are however not recommended for boosting yield in potato rotation regimes. Lablab is a potential crop that can be used in rotation with potato to increase yields and can be recommended to farmers in Nakuru County while rotation of garden pea with potato can be recommended to farmers within Nairobi environs according to the stability studies. Short term evaluation of crop rotations are important predictors to crops that can be used to suppress bacterial wilt effect in farmers fields who are limited by land.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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