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Control of Bacterial Wilt Disease of Tomato Through Integrated Crop Management Strategies

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Tomato cultivation is severely affected by bacterial wilt disease caused by the soilborne pathogen *Ralstonia solanacearum* (previously known as *Pseudomonas solanacearum* Smith). Effects of rotation of tomato (*Lycopersicon esculentum* Mill.) with other crops on soil populations of *R. solanacearum* and on bacterial wilt disease incidence of tomato were evaluated in the field. Monocropped Cassava (*Manihot esculenta* Crantz), *Mucuna puriens* L., *Crotalaria juncea* L., and intercrops of Cassava/*Crotalaria*, Cassava/*Mucuna*, and a natural grass mix (control) were rotated with the tomato cvs. Mira, Ronita, Roma VFN, and Ibadan Local. Monocropped *Mucuna* significantly reduced soil population of *R. solanacearum* by the end of the rotation period, whereas the natural grass rotation had the highest population of the pathogen. Other crops with the exception of monocropped cassava also reduced the pathogen soil population. The incidence of wilt was delayed in cvs. Mira, Roma VFN, and Ronita compared to 'Ibadan Local', but all were 80% or more infected after 8 weeks.

Keywords *Crotalaria juncea*, *Lycopersicon esculentum*, *Manihot esculenta*, *Mucuna puriens*, *Ralstonia solanacearum*, Rotation, Wilt disease.

INTRODUCTION

In addition to their value as a source of vitamins and micronutrients, tomato (*Lycopersicon esculentum* Mill.) is a high-value cash crop. Nigeria is the largest

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tomato producer in West Africa, harvesting 889,900 Mt on about 127,000 ha in 2004 (Food and Agriculture Organization of the United Nations [FAO], 2005). Tomato yields in 2004 were low, average 7.0 Mt·ha^{-1}, compared to average 11.1 Mt·ha^{-1} for all of West Africa (FAO, 2005). Some constraints affecting tomato yields in Nigeria are absence of high-quality seed, suboptimal farming practices, and prevalence of pests and diseases. One of the major diseases of tomato in Nigeria is bacterial wilt, caused by the soilborne pathogen *Ralstonia solanacearum* (Smith) (Adebayo and Ekpo, 2005; Osunde and Ikediugwu, 2002).

The bacterium has a worldwide distribution and a large host range of more than 200 species in 50 families (Hayward, 1991). This is particularly true of race 1, which is especially virulent against solanaceous crops. Epidemics of bacterial wilt disease of tomato have been recorded in Nigeria since 1996 (Adebayo and Ekpo, 2005), leading to restrictions in production in some areas. Survival of this pathogen is particularly encouraged by high soil moisture (Abdullah et al., 1983), which usually typifies major production areas.

Various strategies have been developed for control of bacterial wilt of tomato, but many are limited in application, being site specific or limited by socio-economic conditions (Hayward, 1991). Although host resistance is the most economical control option, it is difficult to obtain cultivars with stable resistance across locations under conditions of high temperature and humidity in the tropics (Hanson et al., 1996). Cultural control strategies using intercropping of non-host crops and soil amendments have been reported (Michel et al., 1997; Sood et al., 1998; Sun and Huang, 1985). Crop rotation, intercropping, or incorporating green manure crops such as sun hemp (*Crotalaria juncea* L.) and mungbean (*Vigna radiata* L.) before planting susceptible crops have been reported to be effective for bacterial wilt disease control (Hartman et al., 1993).

The N requirement of tomato crops in southwestern parts of the country is 60 kg·ha^{-1} of N (Olufolaji, 2000). However, soil nitrogen is usually deficient under continuous cropping, which is a common phenomenon in tomato-based cropping systems in Nigeria (Erinle, 1989). Survival of *R. solanacearum* in the soil is largely dependent not only on the initial population levels in the soil but on the biological, chemical, and physical properties of the soil and the type of cropping system used (Shekhawat and Perombelon, 1991). This determines the availability of host materials, type of debris that can serve as shelter sites, and microbial activity governing antagonistic activities (Hayward, 1991). Apart from the ability of green manure such as *Mucuna* and *Crotalaria* to control bacterial wilt disease (Hartman et al., 1993), they have also been reported to restore and conserve mainly soil nitrogen (Huang and Huang, 1993). Integrated management systems incorporating use of resistant cultivars and appropriate cropping systems can alter the suitability of the microenvironment to the pathogen and influence disease incidence. The objective of this study was to investigate the role of short rotations of cultivars of tomato with other crops on soil populations of *R. solanacearum* and incidence of bacterial wilt disease of tomato.

MATERIALS AND METHODS

The trial was conducted from July 2003 to November 2004 in a naturally infested field at the National Horticultural Research Institute (NIHORT), Ibadan (3°54 E, 7°30 N), in the savannah zone of the southwestern Nigeria. Rainfall is bimodal with an annual mean of 1280 mm; mean minimum temperature range between 20.0 and 22.8°C, and a maximum temperature range between 27.9 and 34.7°C.

The soil pH was 6.35 with a texture of sand (90.8%), silt (1.8%), and clay (7.4%) and with exchangeable Ca 0.99 $cmol{\cdot}kg^{-1}$; Mg 0.38 $cmol{\cdot}kg^{-1}$; K 0.17 $cmol{\cdot}kg^{-1}$; Na 0.4 $cmol{\cdot}kg^{-1}$; exchangeable acidity 0.09 $cmol{\cdot}kg^{-1}$; effective cation exchange capacity (ECEC) 2.03; 96% base saturation; organic carbon, 2.8 $g{\cdot}kg^{-1}$; total N 0.3 $g{\cdot}kg^{-1}$; and available P 9.58 $mg{\cdot}kg^{-1}$.

Cuttings of cassava (*Manihot esculenta* Crantz), cv. TMS30572, and seed of *Mucuna puriens* L., cv. IRZ, and *Crotalaria juncea* L., were collected from the International Institute of Tropical Agriculture (IITA, Ibadan, Nigeria). Tomato seeds were from AGRINOVA (Dallas, Texas), with the exception of 'Ibadan Local', which was from the gene bank of NIHORT.

Effect of Crop Rotation on Soil Population of *R. solanacearum*

The experiment was arranged in a randomized complete block design with three replications. Treatments were monocropped Cassava, *Mucuna*, or *Crotalaria*; intercrops of Cassava/*Crotalaria*, Cassava/*Mucuna*; and a natural grass fallow (control). Each treatment plot was 4 × 4 m in dimension. All rotational crops were planted on 27 Sept. 2003. Cassava was planted using a spacing of 1 × 1 m. The intercrops were planted in alternate rows (1 m apart) such that each plot had two rows of cassava and two rows of either *Crotalaria* or *Mucuna*. Monocropped planting of *Mucuna* and *Crotalaria* consisted of direct drilling in four rows per plot at spacing of 5 cm within row and 1 m between rows. Soil populations of *R. solanacearum* were determined at planting, mid-season (6 months after planting [MAP]) and at harvest of cassava tubers (11 MAP). The total soil nitrogen was determined at the onset of the rainy season, in April 2004, to assess the effect of green manure crops (*Crotalaria* and *Mucuna*) on soil N.

Enumeration of *R. solanacearum*

Five composite soil samples taken at a depth of 0.2 m and weighing approx. 1 kg were obtained with a soil auger in each plot during each sampling period. From those, 10-g subsamples were suspended in 100 mL of sterile distilled water. Tenfold dilutions were prepared from the stock and 0.1 mL of dilutions 10^{-3} and 10^{-4} were transferred to plates containing sterile triphenyl tetrazolium chloride agar. Plates were incubated at 30°C for 48 hours, after which typical fluidal colonies pink at the center and surrounded by a white halo were counted (Kelman, 1954).

Effect of Crop Rotation on Incidence of Bacterial Wilt

After the harvest of cassava (11 Aug. 2004), plots were cleared manually, cutting back all other crops, and divided into four subplots (4 × 1 m). Each treatment plot was planted to four tomato cultivars on 6 Sept. 2004. Four-week-old tomato cultivars (Table 1) previously raised in the nursery in a sand:topsoil:poultry manure mixture (1:2:1 v:v:v) by volume were transplanted at a spacing of 50 × 75 cm. Fertilizer (NPK, 20:10:10) was applied 4 weeks after transplanting at the rate of 60 $kg \cdot ha^{-1}$. Weeding was done manually three times. Incidence of bacterial wilt was monitored fortnightly and expressed as percentage incidence.

Data Analysis

Soil populations of *R. solanacearum* data were transformed using (Log + x) before analysis and arcsine transformation was used for the data on percentage incidence of bacterial wilt of tomato. All data were analyzed using the GLM procedures in SAS (SAS Institute, Inc., Cary, N.C.).

RESULTS

Rainfall patterns during the two years were different (Table 2). The previous non-tomato rotation crop affected soil nitrogen (Table 3). The lowest N content was found in soil under monocropped cassava and natural grass, which were similar. All other treatments were higher and similar.

The previous non-tomato rotation crop affected soil populations of *R. solanacearum* (Figure 1). At establishment, there were no significant differences between treatments in soil populations of *R. solanacearum*, indicating

Table 1: Characteristics of tomato cultivars used.

Cultivar	Source	Growth habit	Reaction to bacterial wilt
Roma	Agrinova Seed	Determinate	Moderately tolerant
VFN	Dallas, Texas	Determinate	Susceptible
Ronita	Agrinova Seed	Determinate	Moderately tolerant
Mira	Agrinova Seed	Indeterminate	Susceptible
Ibadan Local	NIHORT, Ibadan		

Table 2: Rainfall (mm) data, 2003 and 2004.

Month	2003	2004
January	49.1	52.9
February	15.4	124.7
March	22.2	4.9
April	148.6	173.9
May	68.4	151.1
June	378.8	242.4
July	130.9	115.6
August	63.4	68.3
September	286.8	131.9
October	291.2	167.4
November	174.1	7.8
December	0.0	0.0

Table 3: Effect of the initial crop in the rotation on total soil nitrogen at 0–15 cm depth at the onset of the rainy season, April 2004.

Initial crop	Total N ($g \cdot kg^{-1}$)
Cassava monocrop	0.29b[a]
Crotalaria monocrop	0.48a
Mucuna monocrop	0.51a
Cassava + *Mucuna*	0.45a
Cassava + *Crotalaria*	0.37a
Natural grass fallow	0.28b

[a]Means followed by the same letter are not significantly different, $P < 0.05$, Duncans Multiple Range Test.

uniformity in the soil population of the pathogen. By 6 MAP, significant differences were recorded in the soil population of *R. solanacearum*. Monocropped *Crotalaria*, *Mucuna*, and the Cassava/*Mucuna* intercrop had a significantly lower population of *R. solanacearum* compared with the natural grass fallow (Figure 1). At final harvest of cassava, 11 MAP, monocropped *Mucuna* had the lowest population, whereas the natural grass fallow still had the highest soil population of *R. solanacearum*.

Rotation affected bacterial wilt incidence (Figure 2). Irrespective of cultivar, tomato following the natural grass fallow had significantly higher wilt incidence (28.1%) compared with other treatments at 4 weeks after transplanting (WAT). By 6 WAT, wilt incidences of 33.2%, 33.8%, and 34.4% in plots that had Cassava + *Mucuna*, Cassava alone, and *Mucuna* alone, respectively, were significantly lower than other treatments. By 8 WAT, tomato planted in plots previously planted to Cassava + *Mucuna* still had the lowest disease incidence, whereas plants in natural grass fallow plots had the highest wilt incidence (89.4%).

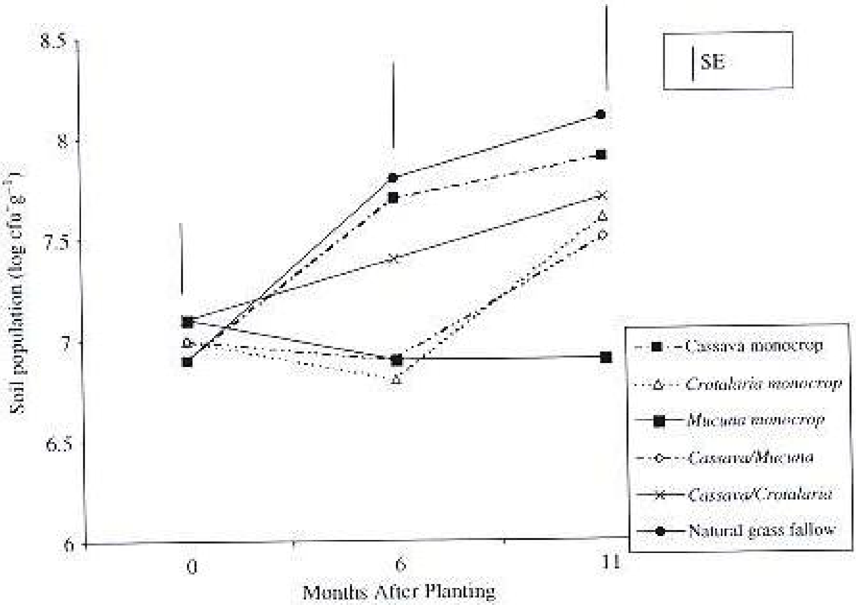

Figure 1: Effect of initial crop on the population of the soilborne pathogen *Ralstonia solanacearum* at various sampling dates 2003, 2004.

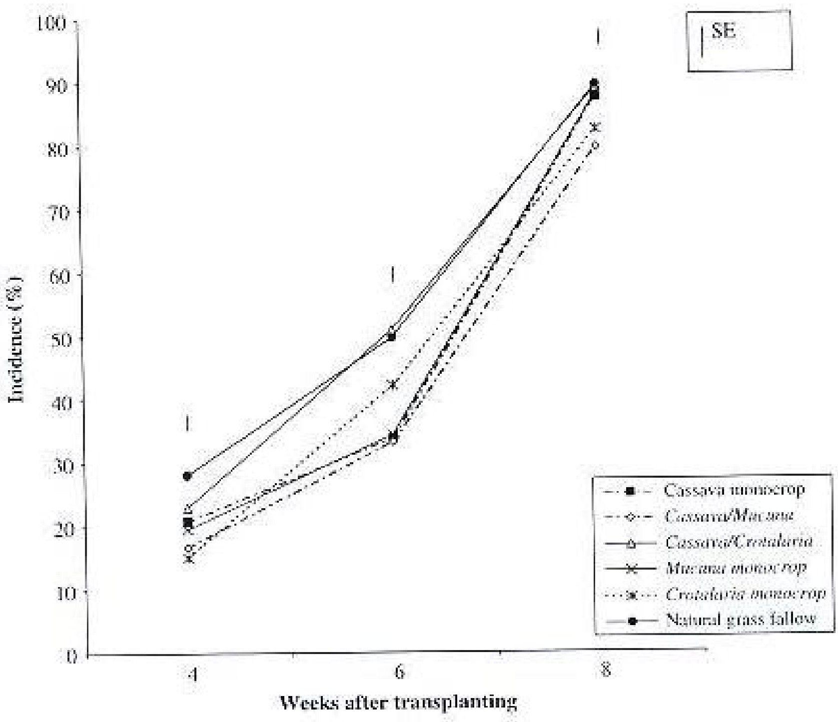

Figure 2: Effect of initial crop on incidence of bacterial wilt of tomato, 2004.

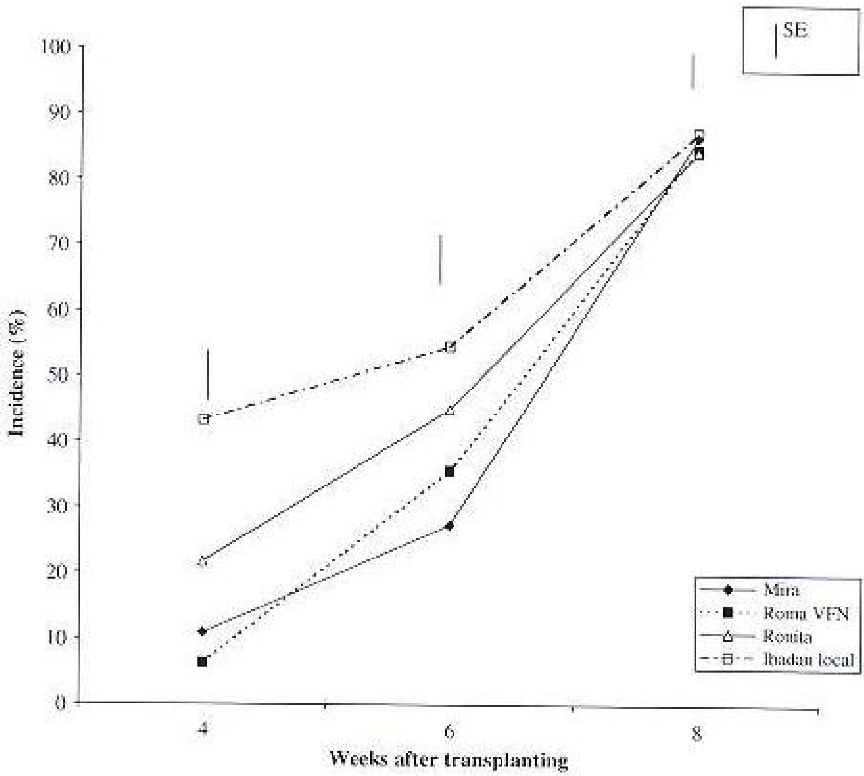

Figure 3: Effect of cultivars on incidence of bacterial wilt of tomato, 2004.

Cultivar affected bacterial wilt incidence (Figure 3). At 4 WAT, irrespective of treatments, 'Roma VFN' had a significantly lower wilt incidence followed by 'Mira', and 'Ibadan Local' had the highest incidence. By 6 WAT, 'Mira' had the lowest incidence, followed by 'Roma VFN'. Wilt incidences in 'Roma VFN' and 'Ronita' were similar and significantly lower than incidences in the other cultivar at 8 WAT. The rotation × cultivar interaction was significant at 4 and 6 WAT but by 8 WAT no significant interaction occurred (Table 4).

DISCUSSION

These studies were undertaken to evaluate the potential of rotating tomato with other crops to reduce soil population of *R. solanacearum* and incidence of bacterial wilt disease. The rotation of tomato with monocropped *Mucuna* was most effective in reducing the soil population of *R. solanacearum* prior to planting tomato. Also, Cassava/*Mucuna* intercrop and monocropped *Crotalaria* reduced pathogen population, whereas monocropped Cassava and the natural grass did not reduce pathogen populations.

Table 4: Incidence of bacterial wilt of tomato as influenced by rotation and cultivar.

Rotation	Cultivar	Incidence (%) 4 WAT[a]	6 WAT	8 WAT
Cassava	Mira	12.0	21.5	89.4
monocrop	Roma VFN	18.6	37.3	89.4
Cassava + *Mucuna*	Ronita	27.1	41.7	81.6
Cassava + *Crotalaria*	Ibadan Local	25.3	34.6	89.4
Mucuna monocrop	Mira	11.5	9.5	84.0
Crotalaria	Roma VFN	0.57	23.6	73.2
monocrop	Ronita	19.6	46.7	84.3
Natural grass fallow	Ibadan Local	35.3	52.9	76.8
Significance of F	Mira	9.1	41.2	89.4
Rotation (R)	Roma VFN	0.57	48.6	89.4
Cultivar (C)	Ronita	37.5	54.3	83.8
R × C	Ibadan Local	45.0	59.4	89.4
	Mira	0.57	11.0	89.4
	Roma VFN	9.1	18.5	83.8
	Ronita	23.6	44.5	89.4
	Ibadan Local	45.1	63.6	89.4
	Mira	0.57	25.9	78.1
	Roma VFN	6.3	48.1	89.4
	Ronita	3.1	39.2	72.1
	Ibadan Local	48.6	55.5	89.4
	Mira	31.6	55.1	89.4
	Roma VFN	0.57	38.1	89.4
	Ronita	19.9	43.9	89.4
	Ibadan Local	60.2	61.8	89.4
		***	***	**
		***	***	NS
		***	**	NS

[a]WAT = weeks after transplanting.
NS, **, ***Not significant, or significant at $P < 0.01$, or $P < 0.001$, ANOVA.

The reason that *Mucuna* reduced the pathogen population is not clear. There have been no reports of a direct bactericidal effect on *R. solanacearum* by *Mucuna*. However, like other green manures, *Mucuna* may restore and conserve N, which could encourage an increase in the biological buffering capacity of the soil and in the process enrich the soil with microflora like bacilli, *Arthrobacter* sp., and *Burkholderia cepacia*, which are competitive with the pathogen (Huang and Huang, 1993; Kloepper et al., 1999). *Crotalaria* sp., on the other hand, has been reported to effectively reduce the pathogen population and control tomato bacterial wilt under screenhouse conditions (Hartman et al., 1993).

Greater attention should be given to using these crops as possible components of an integrated strategy for tomato bacterial wilt control. Natural grass fallow is not a suitable alternative to non-host crop rotation because many weeds serve as alternative hosts for the bacterial wilt pathogen. Seasonal growth of these plants also favors maintenance of the pathogen population in the soil, particularly on wilt-infested land (Hayward, 1991). Similarly, monocropped cassava supported high pathogen populations and appears to confirm the report of Hayward (1991). Bacterial wilt of cassava is prevalent in Indonesia (Nishiyama et al., 1980); it has not been reported on cassava in Nigeria despite the fact that cassava is widely grown in most tomato production areas in the country. This implies that cassava may have been a symptomless host serving as sheltered sites where the bacterium could survive.

The choice of tomato cultivars is clearly an important factor in the success of the management strategy for control of tomato bacterial wilt disease. The introduced cvs. Mira, Roma VFN, and Ronita were least affected early on, whereas the susceptible local landrace was quickly affected, exhibiting high disease incidence even at 4 WAT. There is a need to identify tomato cultivars with higher levels of wilt resistance that are adaptable to tropical conditions.

Disease incidence was still generally high by the end of the growing period. This is likely a result of deliberate choice of high and uniform inoculum at the sites and the highly conducive climatic conditions for bacterial wilt development. Further studies are necessary to identify other complementary crop protection measures that can be introduced to achieve more sustainable control.

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