Compost and *Glomus mosseae* for Management of Bacterial and Fusarium Wilts of Tomato

L. B. Taiwo
D. T. Adebayo
O. S. Adebayo
J. A. Adediran

ABSTRACT. Bacterial and fungal wilts cause considerable yield loss in tomato (*Lycopersicum esculentum* Mill.), and require sustainable control strategies to reduce their incidence. Tomato was inoculated with the arbuscular mycorrhizal fungus *Glomus mosseae* (Nicolson & Gerdemann) Gerd. et Trappe, and treated with organic and inorganic fertilizers to determine effects on severity of tomato wilt caused by *Ralstonia solanacearum* (Smith) and *Fusarium oxysporum* Schlecht. f. sp. *lycopersici* Sacc. (*Fol*) in greenhouse and field studies. In the greenhouse, *Fol* significantly increased wilt of control plants relative to compost-fertilized and *G. mosseae*-inoculated plants. No fruit was produced by inorganically fertilized plants inoculated with *R. solanacearum*, while few fruit were obtained from *Fol*-infected plants. In the field tomato plants fertilized with compost made from cassava peel waste and poultry droppings plus 60 kg NPK had the highest survival of over 55%, while

L. B. Taiwo (E-mail: lbtaiwo@yahoo.com) and J. A. Adediran are affiliated with the Institute of Agricultural Research and Training, Obafemi Awolowo University, P.M.B. 5029, Ibadan, Nigeria.

D. T. Adebayo is affiliated with the Federal College of Agriculture, the Institute of Agricultural Research and Training, Obafemi Awolowo University, P.M.B. 5029, Ibadan, Nigeria.

O. S. Adebayo is affiliated with the National Institute of Horticultural Research, P.M.B. 5432, Idi Ishin, Ibadan.

Address correspondence to: L. B. Taiwo at the above address.

to controls. Application of an additional 60 kg·ha^{-1} of urea to compost significantly decreased survival of tomato in the field. Fertilizer treatment and mycorrhizal inoculation increased the vitamin C content of tomato. *doi:10.1300/J512v13n02_05 [Article copies available for a fee from The Haworth Document Delivery Service: 1-800-HAWORTH. E-mail address: <docdelivery@haworthpress.com> Website: <http://www.HaworthPress.com> © 2007 by The Haworth Press, Inc. All rights reserved.]*

INTRODUCTION

Various insects and diseases stress and damage tomato (*Lycopersicum esculentum* Mill.) plants and reduce yield quality and/or quantity. On tomato an important disease is caused by the bacterium *Ralstonia solanacearum* (Smith), with wilting being the main symptom in the field and greenhouse. Fusarium wilt, caused by the fungus *Fusarium oxysporum* Schlecht. f. sp. *lycopersici* Sacc. (*Fol*) is a significant disease which can also cause plant death. These pathogens are soil-borne and travel through the xylem where vessels are blocked and plants wilt. The endemic nature of wilt diseases can detrimentally affect yield of tomato. According to Sikora and Gazaway (1999) wilt diseases can occur in newly cleared lands, and in areas where susceptible crops have not been previously grown.

Chemical measures have not been satisfactory for control of these diseases. While chloropicrin treatment of a contaminated seedbed gives full season control, methylbromine and methane plus sodium are less effective (Shelf and Macneb, 1986). Well nourished plants are better able to withstand stress. However, application of synthetic fertilizer has not been beneficial for control of bacterial or Fusarium wilts of tomato. Use of resistant cultivars is a viable method for wilt control (Supriadi, 2000). Cultural practices and crop rotation only provide limited control (Kucharek, 1998). Biological control is an attractive alternative to use of synthetic pesticides. Supriadi (2000) reported suppression of tomato wilt caused by *R. solanacearum* using a fluorescent *Pseudomonas* bacterium. It may

be that this is owing to a greater ability of the antagonist to compete for nutrients than do the pathogens, or maybe because of production of lytic enzyme by the antagonist against the pathogen (Waceke, 2002).

Apart from nutrient acquisition for its symbiot, mycorrhizal infection may protect roots from soil pathogens (Akkopru and Demir, 2005). The extent to which the antagonistic organisms suppress or prevent fungal and bacterial attack is uncertain. Dehne (1982) reported that damage due to pathogens was reduced in 17 of 32 reports, while Akkopru and Demir (2005) reported reduction in disease severity because of the presence of *mycorrhizae*. Use of composts to reduce incidence of diseases in crop plants was observed by King (1967). According to Waceke (2002), beneficial organisms can be added directly to the soil, or the soil environment can be made more favorable for them through use of organic amendments.

This project was undertaken to evaluate the impact of compost, with and without amendment with synthetic fertilizer, or mycorrhiza for control of bacterial and fungal wilts of tomato.

MATERIALS AND METHODS

Two glasshouse experiments and a field study were conducted in 2004; and the field study was repeated in 2005 at the Institute of Agricultural Research and Training, Ibadan, Nigeria, located at 70° 56′ N and 30° 45′ at an altitude of 240 m above sea level. In all cases the indeterminate tomato, cv. 'Ibadan Local', obtained from the National Institute of Horticultural Research, Ibadan, was used.

First Greenhouse Study

The treatments consisted of the following: (1) a rate equivalent to 5 Mt·ha^{-1} of composted cassava peel waste and poultry litter (3:1 w/w) prepared as described by Taiwo and Oso (2003), (2) inorganic fertilizer (60 kg·ha^{-1} of urea), with a basal application of phosphorus (P_2O_5) from single super phosphate and potassium (K_2O) from muriate of potash, and (3) a vesicular-arbuscular mycorrhizal fungus, *Glomus mosseae* (Nicolson & Gerdemann) Gerd. et Trappe. A quantity of 30 g per pot of crude inoculum of the soil culture of *G. mosseae* containing root fragments of the trap crop, and mycelium, hyphae, and spores (650-700 spores/ 100 g soil) of the fungus was used as the inoculant. The crude inoculum was introduced into the planting holes of the inoculated pots containing

of attenuated inoculum was applied into the planting hole of uninoculated plots. Unfertilized, uninoculated, plants served as controls. Treatments were arranged in a completely randomized block design with 15-single pot replicates per treatment. Tomato seedlings, 4 weeks old, raised in soil sterilized with electric heat (110°C for 1 h) in a seedling box, were transplanted into 5 L pots filled with field soil sterilized in the same manner. The bacterium *R. solanacearum* was grown on Triphenyl Tetrazolium Chloride (TTC) agar and 30 ml of the pure liquid culture was used. Colonies were washed from the agar surface with sterile distilled water, and centrifuged. The centrifugation and washing procedure was repeated several times. The final population was approximately 10^7 $CFU \cdot mL^{-1}$. The washed inoculum was placed into the soil in pots a week after transplanting. Data on survival and yield were obtained.

Second Greenhouse Study

Fol was grown on potato dextrose agar for 7 days and the culture washed with sterile distilled water to dislodge microconidia. The concentration was adjusted to 10^7 microconidia·mL^{-1}. Inoculation was by placing 30 mL of the conidia suspension in each pot a week after transplanting. Treatments were arranged in a completely randomized block design with 15-single pot replicates per treatment. Each pot had two plants.

Treatments consisted of the following: (1) compost obtained from cassava peel waste and poultry droppings as described previously, (2) inorganic fertilizer at rates described previously, and (3) *G. mosseae* (Nicolson & Gerdemann) Gerd. et Trappe. Unfertilized and uninoculated plants served as controls. Data on survival and yield were obtained.

Field Study

Experiments were conducted in an Alfisol, Iwo series, loamy sand with a history of bacterial and fungal wilts. Pre-cropping soil analysis was done. Soil nitrogen (N) was determined using the micro-Kjeldahl method according to Bremer (1965) while organic carbon determination was by oxidation with sulphuric acid using the method of Wakley and Black (1965). Exchangeable cations were leached from soil with N ammonium acetate before K was determined on a Energy Emitted Lithium flame photometer (Instrumentation Laboratory, Inc., Lexington, MA; AOAC, 1980). Avail-

The soil, to a 15-cm depth contained 12 g·kg^{-1} organic carbon, 1.5 g·kg^{-1} total N, and 4.71 mg·kg^{-1} available P. The exchangeable Na, K, and Ca were 0.29, 0.15, and 3.95 Cmol·kg^{-1} soil, respectively. The levels of nutrients in the soil were of medium status (Sobulo and Adepetu, 1987) requiring a minimal nitrogen application. On April 6, 2005, tomato seeds were sown in a nursery bed. Any existing cover in the field was cut and plowed under. After 14 days the soil was disked and smoothed. Treatments were (1) compost at a rate equivalent to 5 Mt·ha^{-1}, (2) compost + 30 kg urea, (3) compost + 60 kg urea, (4) compost + 30 kg of N:P:K (20:10:10) with an additional basal application of phosphorus (P$_2$O$_5$) from single super phosphate and potassium (K$_2$O) from muriate of potash, (5) compost + 60 kg of N:P:K (20:10:10), and the P$_2$O$_5$ and K$_2$O as before, and (6) *G. mosseae* alone without additional fertilizer. Controls did not receive fertilizer. The compost was incorporated into the soil to a depth of 5 cm and covered with top soil. After a week, 4-week old transplants were established in the field at a spacing of 60 cm × 60 cm. The crude *G. mosseae* inoculum was placed into planting holes of the inoculated plots at transplanting. Half of the inorganic fertilizer N was added a week after transplanting and a second application was made 2 weeks later. Seven treatments were arranged in 10 m × 5 m plots in a randomized complete block design with four replications. Plants were rainfed and staked before flowering. Weeding was done manually and insect pests were controlled with Karate® at the rate of 2 mL·L^{-1} of water every 2 weeks starting 2 weeks after transplanting and continuing until fruit formation.

Data on plant survival, fruit yield, and fruit vitamin C content were obtained. Vitamin C content of fruit was analyzed using the method of Pearson (1987). Data were subjected to analysis of variance to determine involvement of main effects or interactions. If interactions were present they were used to explain results. If not present means were separated using Duncan's Multiple Range Test (Steel and Torrie, 1980).

RESULTS

The compost contained low amounts of N, P, and K, an adequate amount of micronutrients, a negligible heavy metal content, and a high level of microbes (Table 1).

First Greenhouse Study

R. solanacearum: Treatments affected plant survival and yield in the

TABLE 1. Chemical and biological composition of the compost used.

Properties	Value
Organic C (%)	25.0
Total N (%)	1.75
C/N ratio	14.70
NO_3-N (%)	ND[z]
NH_4-N (%)	0.03
P (%)	0.70
K (%)	1.25
Ca (%)	1.47
Mg (%)	0.19
Fe (mg·kg^{-1})	42.0
Zn (mg·kg^{-1})	0.90
Mn (mg·kg^{-1})	1.70
Cu (mg·kg^{-1})	0.52
Cd (mg·kg^{-1})	0.02
Pb (mg·kg^{-1})	0.07
Microbial population (CFU × 10^9)	7.2

[z]ND = Not detected.

TABLE 2. Percentage plant survival and yield of tomato plants infected with *Ralstonia solanacearum* as influenced by fertilizer application and mycorrhizal inoculation in the greenhouse.

Treatment[z]	Percent survival	Number of fruit per plant	Fruit yield per plant (g)
Control	33.0c	3b[y]	36.6b
Compost	87.0a	21a	224.6a
NPK	33.0c	0c	0.0c
Glomus mosseae	50.0b	3b	43.0b
Average	50.8	19.1	18.7

[z]Compost was used at the rate of 5 Mt·ha^{-1}; NPK (20:10:10); number of plants/treatment was 24.
[y]Means within a column followed by the same letter are not significantly different, $P \leq 0.05$, Duncan's Multiple Range Test.

Inoculation with *R. solanacearum* resulted in 50% survival. Compost in-
creased seedling survival 2.6 f

with compost had significantly more fruit and greater yield/plant than for the other treatments.

Second Greenhouse Study

Fol: Treatment affected plant survival and yield (Table 3). Controls and plants receiving NPK fertilizer had the fewest surviving plants and the highest survival was for *G. mosseae* and compost-treated plants. The least number of fruit, and least fruit weight, was produced on plants treated with inorganic fertilizer. Numbers of fruit and fruit yield, for all other treatments were higher and similar.

Field Study

The rainfall pattern was consistent with that typical of precipitation in southwestern Nigeria, and was similar in both years (Figure 1). There was a total of 1,528.1 mm precipitation in 2004 and 1,465.4 mm in 2005. Rainfall in May and July 2005 were higher by 80 and 40% during May and July 2005, respectively, than for the same months during 2004. Fertilizer treatment affected plant survival, number of fruit, yield, and vitamin C content of tomato fruit, and year significantly affected fruit number (Tables 4 and 5).

Almost 84% of control plants were lost to wilt, a value similar to that for plants treated with compost + 60 kg urea (Table 6). The greatest survival was in plots fertilized with compost + 30 kg urea, compost + 60 kg of

TABLE 3. Percentage plant survival and yield of tomato plants inoculated with *Fusarium oxysporum* f. sp. *lycopersici* as influenced by fertilizer application and mycorrhizal inoculation in the greenhouse.

Treatment[z]	Percent survival	Number fruit	Fruit yield (g)
Control	60.0b[y]	33.8a	90.9a
Compost	73.3a	29.0a	85.7a
NPK	66.7b	18.7b	71.5b
Glomus mosseae	76.7a	28.0a	91.1a
Average	69.2	27.4	84.8

[z]Compost was used at the rate of 5 Mt·ha^{-1}; NPK (20:10:10) fertilizer; number of plants/treatment was 30.

[y]Means within a column followed by the same letter are not significantly different, $P \leq 0.05$, Duncan's Multiple Range Test.
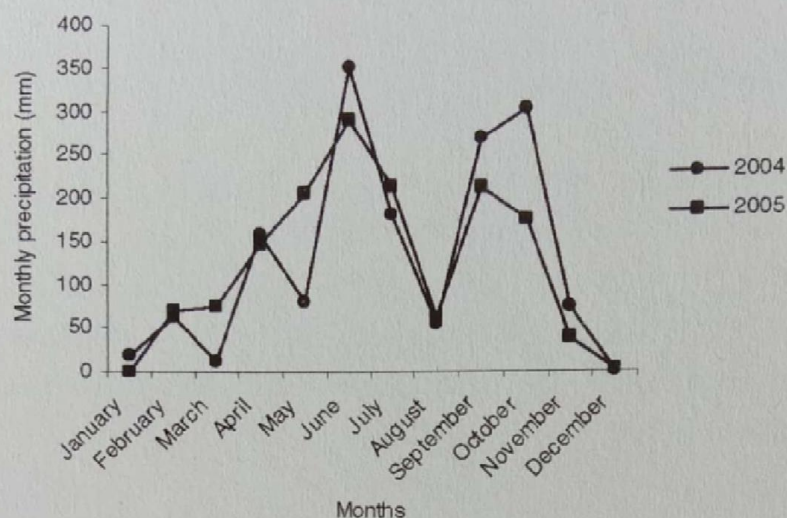
FIGURE 1. Rainfall, 2004 and 2005.

TABLE 4. Analysis of variance table for plant survival and number of fruits for the field experiments.

Source of variation	Number of plants that survived				Number of fruits		
	df	Sum of squares	Mean sum of squares	F	Sum of squares	Mean sum of squares	F
Amendment treatment (T)	6	360.9	60.2	51.9*	87,692.5	14,615.4	5,712.3**
Year (Y)	1	3.5	3.5	3.1	12.1	12.1	4.7*
T × Y	6	4.6	0.8	0.7	16.6	2.8	1.1
Error	28	32.5	1.2		71.6	2.6	

**, *Significant at $P \leq 0.01$ or $P \leq 0.05$, respectively.

TABLE 5. Analysis of variance table for yield and vitamin C content for plants in field experiments.

Source of variation	Yield (kg·ha^{-1})				Vitamin C		
	df	Sum of squares	Mean sum of squares	F	Sum of squares	Mean sum of squares	F
Amendment treatment (T)	6	7.3	1.2	11.5*	542.9	90.5	27.4*
Year (Y)	1	0.3	0.3	3.1	0.2		
T × Y	6						

TABLE 6. Average percent survival and yield of tomato planted in tomato wilt-endemic soil, averaged over years.

Amendment treatment[z]	Percent survival	Fruit yield (Mt·ha^{-1})	Vitamin C content (mg/100 g)
Control	16.5c[y]	0.4b	17.4b
Compost (Cpst)	37.5b	1.8a	27.2a
Cpst + 30 kg Urea	53.5a	1.5a	28.2a
Cpst + 60 kg Urea	21.5c	0.5b	25.6a
Cpst + 30 kg NPK	41.5b	1.3a	24.9a
Cpst + 60 kg NPK	55.0a	1.4a	27.9a
Glomus mosseae	50.0a	1.3a	26.9a
Average	39.4	1.2	25.4

[z]Cpst is compost; NPK was the 20:10:10 compound fertilizer; the number of plants/treatment was 20.
[y]Means within a column followed by the same letter are not significantly different, $P \leq 0.05$, Duncan's Multiple Range Test.

NPK and those treated with *G. mosseae*. The number of fruit was highest in compost only treated plots while the fewest number of fruit were for controls; plots fertilized with compost + 60 kg urea and *G. mosseae*-inoculated plants (Figures 2 and 3). There was no significant difference in numbers of fruit on plants fertilized with compost + 30 kg urea and compost + 60 kg NPK. Fruit yields were generally low (Table 6). The lowest yields occurred in the control plots and plots fertilized with compost + 60 kg urea. The vitamin C content of tomato fruit was least in controls. Vitamin C content of fruit in fertilizer treatments averaged 26.8 mg/100 g.

DISCUSSION

Under greenhouse conditions increased survival of seedlings in treatments that had compost might have been a result of improved soil environment. The low plant survival in controls inoculated with *R. sola-*

FIGURE 2. Effect of organic and mineral fertilizers providing 30 and 60 kg·ha^{-1} of urea and 30 and 60 kg·ha^{-1} of NPK, and mycorrhizae on number of fruit, 2004.
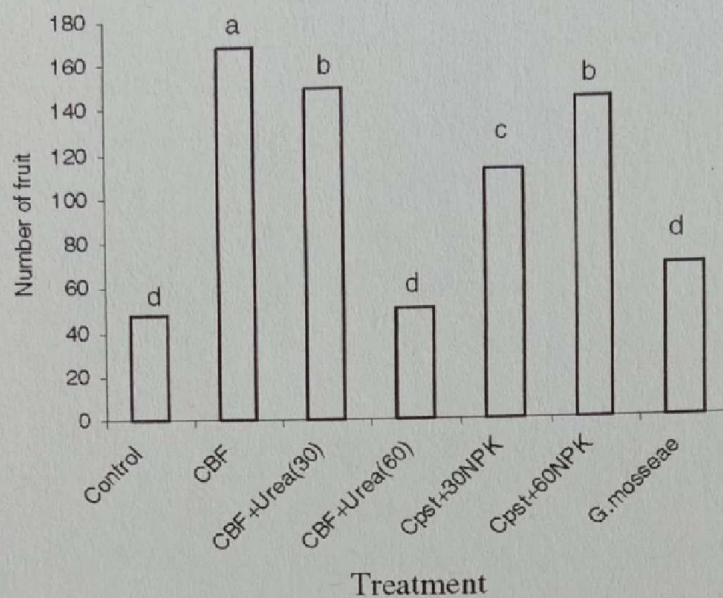
CBF = Compost-based fertilizer. Bars with same letter are not significantly different, $P \leq 0.05$, Duncan's Multiple Range Test.

FIGURE 3. Effect of combined use of organic and mineral fertilizers providing 30 and 60 kg·ha^{-1} of urea and 30 and 60 kg·ha^{-1} of NPK, and mycorrhizae on numbers of fruit, 2005.
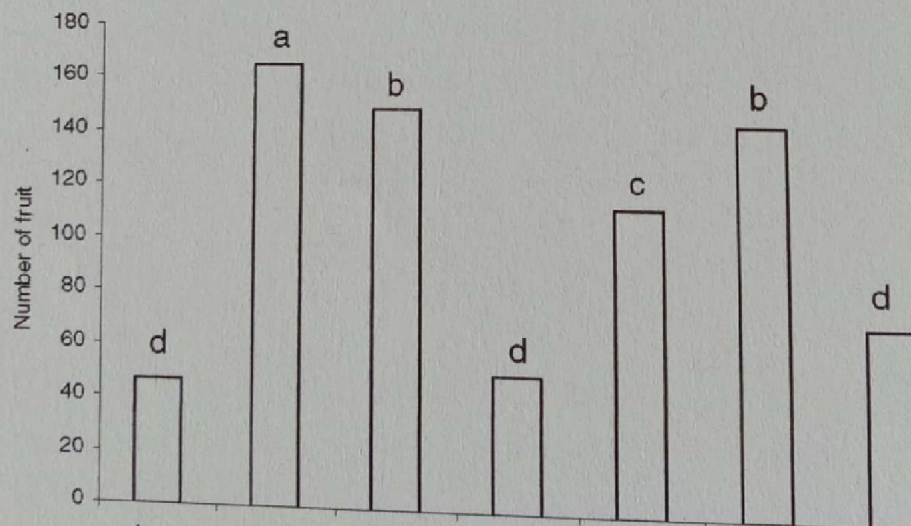
Akkopru and Demir (2005) reported that the soil environment could be made favorable for microbial antagonists through use of compost and other organic amendments. Increase in moisture content of compost from 40 to 50% was found to encourage bacteria and fungal colonization of plants, and increased their disease suppressive capabilities (Hoitink, Stone and Han, 1997). They also reported disease severity was reduced by 8.6-58.6% due to inoculation with *G. intraradices* (Schenk & Smith), and it was noted that inoculation with an individual bacterium was more effective than inoculation with a single arbuscular mycorrhiza, or with a combination of *G. intraradices* + Rhizobacteria. Competition and secretion of antibiotics by some beneficial organisms, and direct parasitism by others, can impede development of pathogens (Noble and Coventry, 2005). Beneficial microbes scavenge nutrients for plants to use. In return, the plant provides carbon in the form of sugars and proteins. This symbiotic system supports the beneficial organisms and the plant, but generally excludes pathogens. Improved phosphorus uptake in the host plant has been associated with mycorrhizal fungi (Waceke, 2002). When plants are not deprived of nutrients, they are better able to tolerate or resist disease-causing organisms (Noble and Coventry, 2005).

In the greenhouse, *Rhizoctonia*-inoculated plants treated with inorganic fertilizer did not produce fruit; additional N from the synthetic fertilizer might have been used by pathogens as a source of nitrogen, allowing increased growth and infectivity. It could also be that the nitrogen caused plants to develop vegetatively and hindered fruit development. In most cases increases in survival of plants in the field, from use of compost alone, and *G. mosseae* relative to controls suggests that compost might affect the pathogens. Compost might have supplied increased levels of nutrients. When combined with urea or NPK survival was generally further improved. Some nutrients have been found to reduce growth of pathogens. High potassium levels retard *Fusarium* in tomatoes (Foster and Walker, 1947). However, a detailed study of the competition between the two microorganisms in the laboratory is required to identify the specific interaction between competing microbes, and the effect of these interactions on the growth and development of pathogens.

Low yields of tomato in the field were a result of low survival. Large

nutrient imbalance resulting from increase in ammonial N. The ammonia form of nitrogen increases disease severity in tomato (Noble and Coventry, 2005) while use of nitrate nitrogen, in soil with a high pH, results in better wilt control in tomato (Woltz and Jones, 1973). Increased vitamin C content in fruit in treated plots relative to control, indicates that the vitamin C level can be increased in tomato with organic or mineral fertilizers.

Generally, percent survival of compost-treated plants was high in both greenhouse and field relative to control; this was not however, translated to increased yield in the greenhouse when the crop was infected with *Fol*. Use of compost alone, or when mixed with inorganic fertilizer for tomato production may be a viable strategy for suppression of *R. solanacearum* or *Fol*. Use of *G. mosseae* also appeared to reduce incidence of wilt of tomato. The impact of *G. mosseae* was not clearly beneficial to fruit yield in the glasshouse when compared with results in the field, which might be owing to there being no restriction in nutrient uptake in soil in the field as opposed to the glasshouse where there was a limited quantity of soil. However, use of *G. mosseae* as a biological control agent requires further study involving higher numbers of spores to ensure a more aggressive level of competition between *G. mosseae* and the wilt-causing pathogens.

LITERATURE CITED

Akkopru, A. and S. Demir. 2005. Biological control of Fusarium wilt in tomato caused by *Fusarium oxysporum* f. sp. *lycopersici* by AMF *Glomus intraradices* and some Rhizobacteria. J. Pathol. 153:544-548.

Association of Official Analytical Chemists (AOAC). 1980. Official methods of Analysis, 11th ed., AOAC, Washington, DC.

Bray, R. H. and L. T. Kurtz. 1954. Determination of total, organic and available forms of P in soil. Soil Sci. 59:39-45.

Bremer, J. M. 1965. Total nitrogen, pp. 119-178. In: C. A. Black (ed.). Methods of soil analysis, Part 2. American Society of Agronomy Vol. 9, Madison, WI.

Dehne, H. W. 1982. Interactions between vesicular mycorrhiza fungi and plants. Plants Path. 72:1115-1119.

Foster, R. E. and J. C. Walker. 1947. Predisposition of tomato to Fusarium wilt. J. Agricult. Rsch. 74:165-185.

Hoitink, H. A. J., A. G. Stone, and D. Y. Han. 1997. Suppression of plant diseases by composts. HortScience 32:184-187.

Jackson M. J.

Kucharek, T. 1998. Bacterial wilt of row crops in Florida. University of Florida, IFAS, Cooperative Ext. Serv., Circ-1207.

Noble, R. and E. Coventry. 2005. Suppression of soil-borne plant diseases with composts: A review. Biocont. Sci. Technol. 15:2-20.

Pearson, D. 1987. Fruit and vegetable products, pp. 190-226. In: H. Egan, R. S. Kirk, and R. Sawyer. (eds.). Chemical analysis of foods, 8th ed. Longman Scientific and Technical, London.

Shelf, A. F. and A. A. Macneb. 1986. Vegetable diseases and their control. Longmann, Harlow, Essex, England.

Sikora, E. J. and W. Gazaway. 1999. Wilt diseases of Tomato. (available on line at: www.aces.edu).

Sobulo, R. A. and J. A. Adepetu. 1987. Soil testing and fertilizer formulation for crop production in Nigeria. National Fertilizer Seminar, Port Harcourt, October 28-30, 1987.

Steel, R. and J. Torrie. 1980. Principles and procedures of statistics. Abiometric approach, 2nd ed. McGraw-Hill, NY.

Supriadi, R. 2000. Bacterial wilt disease of medicinal plants caused by *Ralstonia solanacearum* and strategy to control the disease. J. Penelitian Pengembangan Pertanian 9:17-26.

Taiwo, L. B. and B. A. Oso. 2003. Influence of composting techniques on microbial succession, temperature and pH in a composting municipal solid wastes. African J. Biotech. 3(4):239-240.

Waceke, J. W. 2002. Use of arbuscular mycorrhiza fungi for improved crop production in sub-saharan Africa. J. Tropical Micro. 1:14-21.

Wakley, A. and C. A. Black. 1965. An examination of the Degtjareff method for determining soils organic matter and a proposed modification of the chromic acid titration method. Soil Sci. 37:29-38.

Woltz, S. S. and J. P. Jones. 1973. Tomato Fusarium wilt control by adjustments in soil