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THE ROLE OF COW DUNG AND KITCHEN MANURE COMPOSTS AND THEIR NON-AERATED COMPOST TEAS IN REDUCING THE INCIDENCE OF FOLIAR DISEASES OF *Lycopersicon esculentum* (Mill)

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Abstract

Compost teas are fermented watery extracts of composted materials used for their beneficial effect on plants. A study was conducted in the field to compare the efficacy of cow dung and kitchen manure composts and their derived non-aerated compost teas on disease symptoms expression and severity of *Lycopersicon esculentum*. The experimental layout was a complete randomised block design comprising six treatments, each of which was repeated three times: the negative control plot (Tm-); the positive control or fungicide plot (Tm+); the cow dung compost plot (Cpi); the kitchen manure compost plot (Cpii); the compost tea derived cow dung plot (Tci); and the compost tea derived kitchen manure plot (Tcii). Compost tea derived cow dung was revealed to be richer in elemental nutrients (N, P, K) than compost tea from kitchen manure, and significantly ($p < 0.0001$) enhanced fruit yield per plant. Similarly, the two composts and their derived compost teas significantly ($p < 0.0001$) reduced the incidence and severity of disease symptoms compared to the controls, with the highest efficacy accounting for cow dung compost and compost tea. Although the non-aerated compost teas were not amended with micro-organisms, these results suggest that the two compost teas in use were rich enough in microbial pathogen antagonists, and therefore, are perceived as potential alternatives to synthetic chemical fungicides. Future work will attempt to identify these microbial antagonists with highly suppressive activity in the non-aerated compost teas.

Keywords: Compost, Compost Tea, *Lycopersicon esculentum*, Fungicide, Disease Symptoms

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Introduction

In Cameroon, agriculture represents up to 20% of the national internal raw products (Anonymous, 2008). The total area under tomato production in tropical Africa is about 300,000 ha with an estimated annual production of 2.3 million tons (Van der Vossen *et al.*, 2004). In Cameroon, Adamawa is the third national producing region, with 51 700 tons after west and centre (AGRI-STAT, 2010). The fruit is rich in vitamins A, C, thiamine, riboflavin, and niacin as well as some minerals like potassium and sodium (Janes, 1994). Moreover, lycopene content in tomato is higher than in other fruits or vegetables, and at least 85 % of its input into the human body comes from tomato or tomato derived products (Bramley, 2000; Chalabi *et al.*, 2004). Tomato culture thus constitutes an income generation activity for many of the growers in rural and urban areas. However, its production is affected by numerous diseases which are the main causes of decrease or total yield losses (Welke, 2005). The present day agriculture is not more sustainable in most parts of the country, because of the dependence of growers on chemical

fertilizers and pesticides from the North. The powerful message that derives from all thoughts and dialogues is to move towards organic farming systems, and/or biological agriculture. Farming systems attempt to provide a balanced environment, in which the maintenance of soil fertility and the control of pests and diseases are achieved by the enhancement of natural processes and cycles, with moderate inputs of energy resources, while maintaining an optimum productivity. Uncontrolled use of chemical agricultural techniques have resulted in a great increase in productivity, but at the cost of negative impacts including soil degradation, resistance to pesticides (Barbier and Catin, 1994), detention of soil health and environmental pollution (Lachance and Rouleau, 2004). Some pesticides or contaminants may accumulate in fruits and tubers (Sonchieu, 2002). As an answer to these problems, recent research has favoured the organic farming with uncontaminated quality compost (Znaïdi, 2002; Ngakou *et al.*, 2008), from which compost tea can be extracted. Compost tea represents not only a mean of

completing the role of compost in the fertilization process, but also a strategy of cultivation of useful microflora to prevent stem and floral diseases (Deschênes, 2007). Hence, there is growing interest in the potential for using composts tea to stimulate root and vegetative growth, prevent and control diseases in field crops, increase crop yields and quality. Information concerning its use and effectiveness is slowly increasing (Litterick *et al.*, 2004; Haggag and Saber, 2007). Previous work conducted in the region of Adamawa focused on the relative effects of compost and non-aerated compost tea on *Solanum tuberosum* production (Ngakou *et al.*, 2012). The chemical components were not assessed. Considering the role of compost tea that depends on the source of composting material and its chemical composition, this research is attempted to assess and evaluate its potential as biological control agent against tomato disease symptoms in the field. The outcomes of this study will enable making up our mind on whether compost tea could be used as a potential substitute of chemical fertilizers and pesticides.

Materials and Methods

Experiments were carried out at Dang-Ngaoundéré in the Adamawa region (Cameroon) located at 7°25'119"N, 13°33'415"E at 1106 m altitude above the sea level. During the field work period from June to October 2011, the average daily rainfall, temperature and relative humidity were 228.14mm, 28.24°C and 80.44%, respectively, with a mean sunlight of 139.22 h per day (CMSN, 2011).

Biological and chemical materials

Seeds of tomato (*Lycopersicon esculentum*) of the « Rio grande » variety were purchased from a phytosanitary stores in the local market. Compost made separately from cow dung and kitchen manure was produced by the research team of the corresponding author as described by Ngakou *et al.* (2008). Compost tea was extracted (Fig. 1) as described by Ngakou *et al.* (2012) from a mixture of compost/tap water in a 1/15 (Kg/L) ratio. After extraction, compost teas were stored in 1.5 L bottles at 4°C in the refrigerator (Fig. 2), and applied to the field at a rate of 5L/plot.

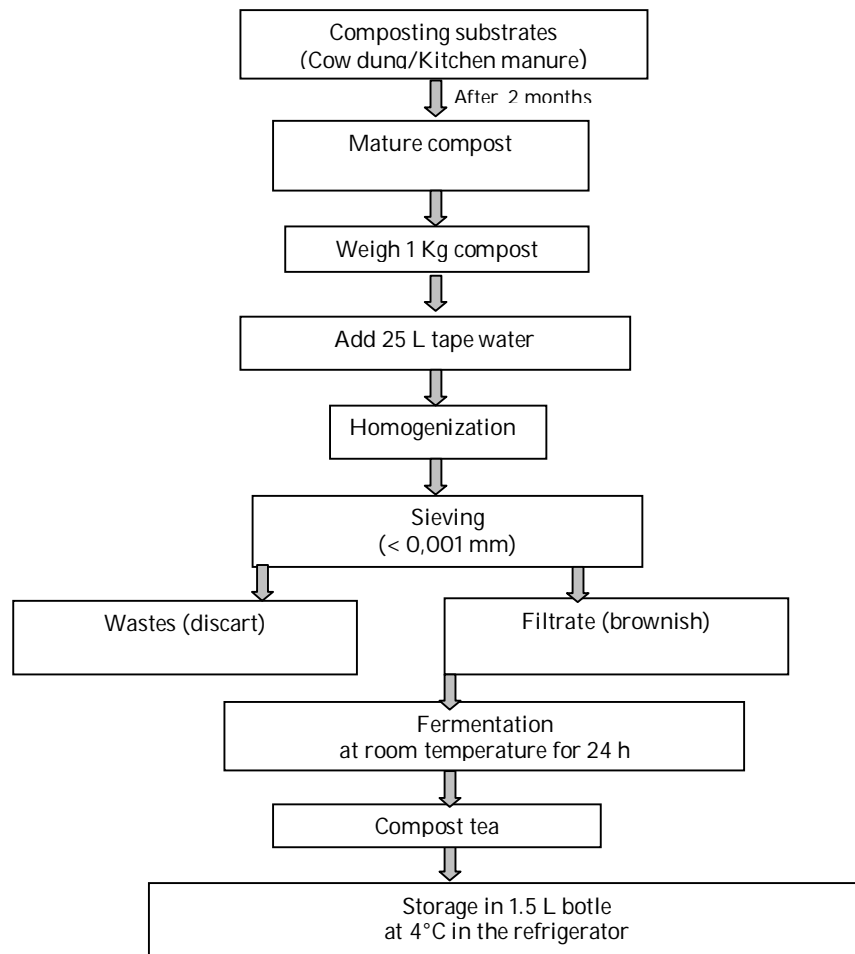


Fig. 1. Extraction process of non-aerated compost tea from a compost



Fig. 2. Sample of compost tea from cow dung (A) and kitchen manure (B)

The fungicide used as positive control was the so called «Fongistar 72% WP», commonly used by tomatoes growers and available in local phytosanitary shops. The fungicide Fongistar 72% WP (25 g) was dissolved in 16 L tap water and applied at a rate of 1.5 L per elemental unit.

Pretreatments and rearing of plantlets

Direct sowing of tomato seeds is not often recommended, since the germination of seeds and health of plantlets is not always guaranteed. Hence, plantlets were reared through a nursery. In the nursery, three plots of 2 m² each were prepared as pretreatments:

Cd: plot covered with 2cm layer of cow dung compost,

Km: plot covered with 2 cm layer of kitchen manure (Km) compost
Tm-: negative control plot covered with 2cm layer of top soil.

Experimental design and treatments

The following 6 treatments were investigated (Table 1). An experimental unit repeated thrice for a treatment was 4.60 x 1.40 m² plot. Each plot contains 20 plants, organized in two rows, and separated 0.40 m apart. The distance from plant to plant in each row was 1 m. Thus, the experimental field was made up of 18 plots, arranged in randomized complete block design (RCBD).

Table 1. Treatments and their significances

Tm+	Positive control derived from Tm- nursery plantlets, to which fungicide was applied at a rate of (25 ml/plantlet)
Cpi	Plots on which each plantlet from Cd nursery pre-treated plants received 50g of Cd compost at transplanting
Cpii	Plots on which each plantlet derived from Km nursery pre-treated plants received 50g of km compost at transplanting
Tci	Plots on which each plantlet derived from Cd nursery pre-treated plants received thrice Cd compost tea at a rate of 50 ml/plantlet after every two weeks as from transplanting
Tcii	Plots on which each plantlet derived from Km nursery pre-treated plants received thrice Km compost tea at a rate of 50 ml/plantlet after every two weeks as from transplanting

Chemical analysis of compost tea

Total Nitrogen: mineralization with Kjeldahl method (AFNOR, 1984), nitrogen determination according to Hantzsch reaction (Devani *et al.*, 1989). Total Phosphorus: as described by Rodier (1978). Total potassium: atomic absorption flame photometer (Electrothermal AAS, Dortmund, Germany) after 1/100 dilution of sieved extracts and spectrophotometric lecture (Elmer, 1994).

Evaluation of growth parameters

Tomato survival rate after transplantation was evaluated per treatment at 7 and 21 days after planting (number survived/number transplanted expressed in %). The height of 10 tomato plants per plot was assessed from the soil level to the apex at flowering using a graduated ruler. The dates of 50% flowering and fructification were also determined.

Determination of disease symptoms

The disease symptoms expressed by tomatoes were assessed at 7, 21 and 35 days after transplantation based on counts of the number diseased plants per treatment and symptoms observed on plant leaves and stems. At 65 days after transplantation, the number of diseased fruits was also assessed by counts on 10 plants per plot. Main symptoms per treatment were assessed, and rated according to their frequencies on plant parts.

Yield assessment

Tomato yield was determined by counting and weighing fruits collected from 10 plants randomly selected plants per elementary unit and per treatment.

Statistical analysis

Data were statistically analyzed by Analysis of Variance (ANOVA) using the Statistical Package for the Social Science (SPSS) program, version 10.0. Means were compared between treatments using the Least Significant Difference (LSD) procedure. Correlations between parameters were assessed using the same SPSS program.

Results and Discussion

Chemical composition of tea

The compost tea was subjected to chemical analysis for the elements nitrogen (N), phosphorus (P) and potassium (K), which are the most requested for plant growth. Results analyzed by ANOVA indicate that the concentrations of N, P and K in cow dung (Cd) compost tea (Tci) were greater than those of Kitchen manure (Km) compost tea (Tcii) treatments, although no significant difference was found for potassium (Table 2). Such elevated concentrations were recently reported from anaerobic compost tea used to control gray mold of tomato with 10.9 mg/L for N, 72 mg/L for P and 421 mg/L for K (Koné *et al.*, 2010). The differences between elemental concentrations of compost tea were previously justified by the variation of the efficacy of compost extracts with both the extraction procedure, the nature of the starting substrate, the duration of the composting process, and the maturity of compost (Brinton, 1995).

Table 2. Elemental composition (N, P, K) of cow dungs and kitchen manure derived compost teas

Compost teas	Concentration of elements (mg/L)		
	N	P	K
Tci	14.49a	26.3a	116.0a
Tcii	12.46b	23.53b	101.2a
LSD	5.03	0.11	ns

For each elemental component value of the same column affected by the same letter are not significantly different at the considered level of probability; Tci: Cow dungs compost tea; Tcii: Kitchen manures compost tea.

Effect of treatments on survival of tomato plants

Nursery plants transplanted to the experimental units were assessed for their ability to survive under different treatments (Fig. 3). Data from this figure show that at 7 days after planting (DAP), the number of plants surviving the chemical fertilizer treatment (Tm+) was slightly but significantly ($p = 0.004$) lower (18) than that of the other treatments (all 20 survived). Twenty-one DAP, the numbers of plants surviving the negative (Tm-) and positive (Tm+) control treatments were significantly ($p = 0.034$) lower compared to those of compost treatments (Cpi

and Cpii). Although the numbers of plants surviving the compost tea treatments (Tci and Tcii) were higher than those of the control, it was not enough to display significant difference. The decrease in the number of control plants from the 7th to the 21st DAP would signify that plants surviving until day 7 DAP died in the following two weeks until 21 DAP, meaning that compost and compost tea contributed to increase the survival of the plants. In a related trial conducted on *Solanum tuberosum*, compost was reported to positively influence by 80% the development of potato plants (Ngakou *et al.*, 2012).

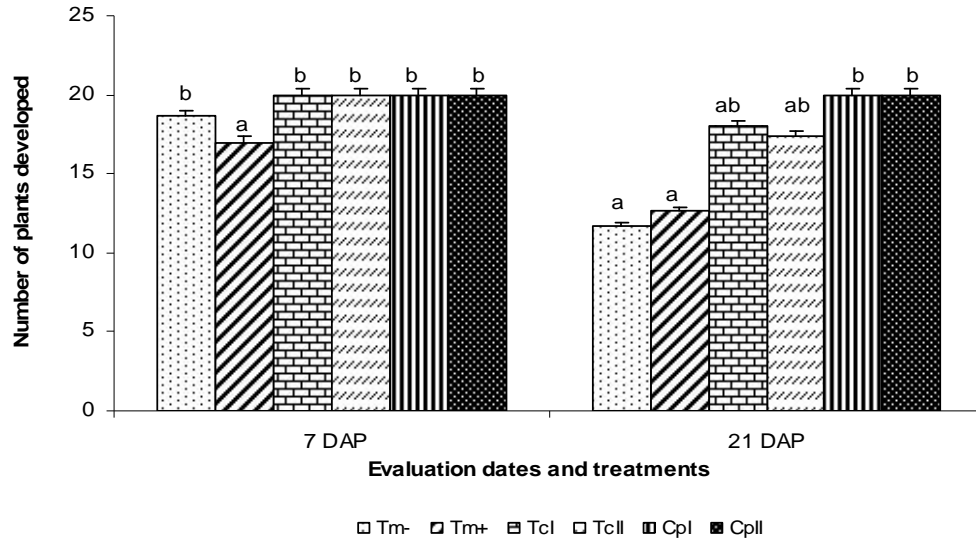


Fig. 3. Effect of compost and compost tea on the development of tomato plants

For each planting date, bars affected by the same letter are not significantly different at the p-value considered. Tm-: Negative control plot on which no compost or fungicide was provided to plantlets; Tm+: Positive control plot on which only a fungicide was provided to plantlets at a rate of 25 ml/plantlet; Cpi: Plot on which each plantlets received 50 g of Cd compost at transplanting; Cpii: Plot on which each plantlet received 50g of Km compost at transplanting; Tci: Plot on which each plantlet received thrice Cd compost tea at a rate of 50 ml/plantlet after every two weeks as from transplanting; Tcii: Plot on which each plantlet received thrice Km compost tea at a rate of 50 ml/plantlet after every two weeks.

Similarly, among the different compost types tested on tomato in the field, compost from horse dung was revealed to be able to stimulate the germination of seeds and development of plant roots (Levy and Taylor, 2003). Compost has been reported to be a best amendment for tomato

plant growth (Meunchang *et al.*, 2006), whereas for other authors, compost tea from animal could be better than compost from plants as growth stimulator (Weltzien, 1991; Al-Dahmani *et al.*, 2003; Haggag and Saber, 2007).

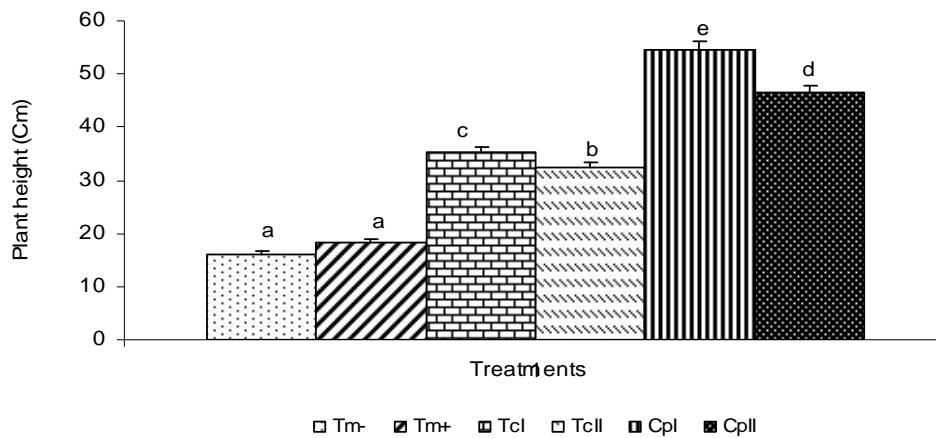


Fig. 4. Height of tomato plants at flowering as influenced by treatments

For each planting date, bars affected by the same letter are not significantly different at the p-value considered. Tm-: Negative control plot on which no compost or fungicide was provided to plantlets; Tm+: Positive control plot on which only a fungicide was provided to plantlets at a rate of 25 ml/plantlet; Cpi: Plot on which each plantlets received 50 g of Cd compost at transplanting; Cpii: Plot on which each plantlet received 50g of Km compost at transplanting; Tci: Plot on which each plantlet received thrice Cd compost tea at a rate of 50 ml/plantlet after every two weeks as from transplanting; Tcii: Plot on which each plantlet received thrice Km compost tea at a rate of 50 ml/plantlet after every two weeks.

Changes in plant heights at flowering under different compost and compost tea receipts

As far as the height of plants is concerned (Fig. 4), the positive and negative control treatments did not have an effect on this parameter. However, composts as well as Cd and Km compost teas significantly increased the height of tomato plants, more than that of the controls, but with a more pronounced effect of compost over the compost tea treatments. Ngakou *et al.* (2012) revealed 80.5% increased of *Solanum tuberosum* height at 65 DAP as the response of spray of plants with compost tea. Compost tea applied on *Vigncola* was also reported to reduce by 60% the stem diameter lesions caused by *Plasmopara viticola*, thus allowing increase in vertical growth of plants (Larbi, 2006). This height improvement of tomato plants by compost could be explained by provision of assimilable nutrients to plants and reduction of the soil acidity through buffer effect (Santerre, 1999). Compost tea activity may reflect the quality of the substrate used, as well as the production process (Brinton, 1995). Its active components (bacteria, fungi) ensure the protection of plants against pathogens, thus favouring its development (Deschênes, 2007). These observations over the controls could be attributed to the beneficial effects of compost on soil fertility, in addition to accumulation of organic carbon (Zinati *et al.*, 2001), that is able to positively influence the plant growth (Chen *et al.*, 1994).

Variation of fruit yield as affected by different treatments

Values estimating the number of fruits revealed a significant difference between treatments (Table 3). Compost and compost tea significantly ($p < 0.0001$) contributed to increase the healthy fruits number compared to the control treatments, the best contribution accounting for Cd compost. Conversely, the negative control significantly ($p < 0.0001$) increased the number of diseased fruits/plant (56%) more than any other treatment, with the best-reduced number of diseased fruit/plant accounting for Cd derived compost tea (95.9%).

Cow dung's compost ranked first in the increment of fruits yield/plant with 26 tomato fruits/ plant, among which 23 were healthy and 3 diseased, whereas the lowest fruit yield was attributed to the negative control treatment, with a total amount of 2 fruits/plant thus 1 healthy and 1 diseased. The two compost tea increased the number of healthy fruits by 84.26% compared to the diseased fruits (15.74%). Kitchen manures compost was revealed to enhance by 95.7% the yield of *Solanum tuberosum* tuber (Ngakou *et al.*, 2012). Compost tea may act within the nearest environment of the plant roots as a protection layer against pathogens, but also as the interface where minerals are uptaken by the host plant roots (Deschênes, 2007).

Table 3. Status of fruits per plant as influenced by compost and compost tea at 65 DAP

Treatments	Healthy fruit/plant	Diseased fruit/plant	Total fruits/plant
Tm-	1.30a	0.73a	2.3a
Tm+	3.40ab	0.36a	5.76ab
Tci	6.50b	0.40a	8.90b
Tcii	5.73b	0.90a	7.63b
Cpi	23.26d	2.93b	26.20d
Cpii	10.96c	3.50b	14.46c
p-value	$p < 0.0001$	$p < 0.0001$	$p < 0.0001$
LSD	4.43	2.03	4.60

Values of the same column affected by the same letter are not significantly different at the p-value considered.

Tm-: Negative control plot on which no compost or fungicide was provided to plantlets; Tm+: Positive control plot on which only a fungicide was provided to plantlets at a rate of 25 ml/plantlet; Cpi: Plot on which each plantlets received 50 g of Cd compost at transplanting; Cpii: Plot on which each plantlet 50 g of Km compost at transplanting; Tci: Plot on which each plantlet received thrice Cd compost tea at a rate of 50 ml/plantlet after every two weeks as from transplanting; Tcii: Plot on which each plantlet received thrice Km compost tea at a rate of 50 ml/plantlet after every two weeks.

The negative and positive control plants were the most affected by fruit looses, with respectively 86.95% and 40.97% damaged fruits. In fact, plants treated with fungicide are protected against a wide range of pathogens, while the negative control plants are unable to ensure this protection. It is then obvious to count more diseased fruits on negative control plants. The application rate of compost tea is very important

since it can contribute, not only to reduce pathogen attacks, but also increase crop yield (Hoikink and Grebus, 1994). A weekly application was shown to decrease the incidence of *Botrytis* on latus, leading to a substantial reduction disease severity and increase of healthy and commercial latus (McQuilken *et al.*, 1994). As far as compost treatment is concerned very few diseased fruits were observed.

During the composting process, a population of antagonistic microorganisms is also developed, and confers to compost the potential to protect plant against diseases or pathogen attacks (Scheuerell and Mahaffee, 2002).

Plant symptoms analysis

Symptoms as appeared on plant parts were recorded at 7, 21 and 35 DAP for each of the treatments (figure 5). Plants showing symptoms at 7 DAP from treatment Cpi, Cpii, Tci, Tcii and Tm+ were significantly ($p = 0.030$) lower in number than those of the negative control (Tm-), although compost was more efficient in reducing disease symptoms on plants than any other treatment.

Whereas the number of symptomatic plants amended with compost was maintained stable at 21 and 35 DAP, plants sprayed with Cd and Km compost teas were significantly ($p = 0.006$) reduced with time, while the number of diseased plants from control treatments were instead enhanced. These results are in agreement with findings of Znaïdi (2002), Deschênes (2007), who reported inhibition or suppression of disease symptoms by compost and compost tea based on competition between the active micro-organisms of compost tea and their plant pathogen antagonists. Compost teas have been revealed to

be natural inhibitors of plant diseases, due to beneficial micro-organisms or active chemical substances they contain (Siddiqui *et al.*, 2008), with animal derived compost tea being more efficient than kitchen manures compost tea (Haggag and Saber, 2007). Conversely, other authors believe that compost types have no direct influence on compost tea, and thus, compost tea from both sources may act on plant pathogen with the same efficiency (Scheuerell and Mahaffee, 2006). However, the results from this study have clearly demonstrated that cow dung's derived compost tea was more effective in reducing tomato disease symptoms than kitchen manures compost tea (confer Fig. 5 at 35DAP). The mechanisms by which compost tea control plant pathogens have been reported to be through beneficial microorganisms, either by their ability to compete with plant pathogens for space and nutrients (Al-Mughrabi *et al.*, 2008), for available food supply, the so called "general suppression or microbiostasis" (Chen *et al.*, 1987; Boehm and Hoikink, 1992), through destruction of the pathogen by parasitism (El-Masry *et al.*, 2002), influence on the phyllosphere (Larbi, 2006), and/or by induction of systemic resistance to plant pathogens (Zhang *et al.*, 1998).

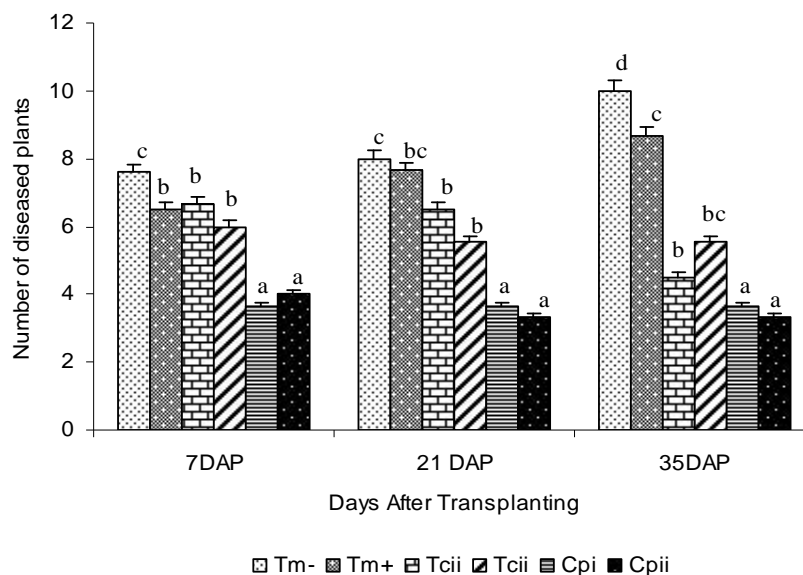


Fig. 5. Number of symptomatic plants per treatment

For each planting date, bars affected by the same letter are not significantly different at the p-value considered
 Tm-: Negative control plot on which no compost or fungicide was provided to plantlets; Tm+: Positive control plot on which only a fungicide was provided to plantlets at a rate of 25 ml/plantlet; Cpi: Plot on which each plantlets received 50 g of Cd compost at trans-planting; Cpii: Plot on which each plantlet received 50 g of Km compost at transplanting; Tci: Plot on which each plantlet received thrice Cd compost tea at a rate of 50 ml/plantlet after every two weeks as from transplanting; Tcii: Plot on which each plantlet received thrice Km compost tea at a rate of 50 ml/plantlet after every two weeks.

Links between observed symptoms and diseases

This was done by observations and counting of diseased plants by determining the individual symptoms appearing on plant leaves, stem, fruits and collar for each treatment at 7 and 21 DAP.

Symptoms identified were foliar and stem spots, apical and total shedding of leaves, stunted of plants, spoilage at the bottom of fruits, black spots on fruits, fissure on fruits, apical shedding, yellowish of leaves, and stem spots/necrosis (Fig. 6).

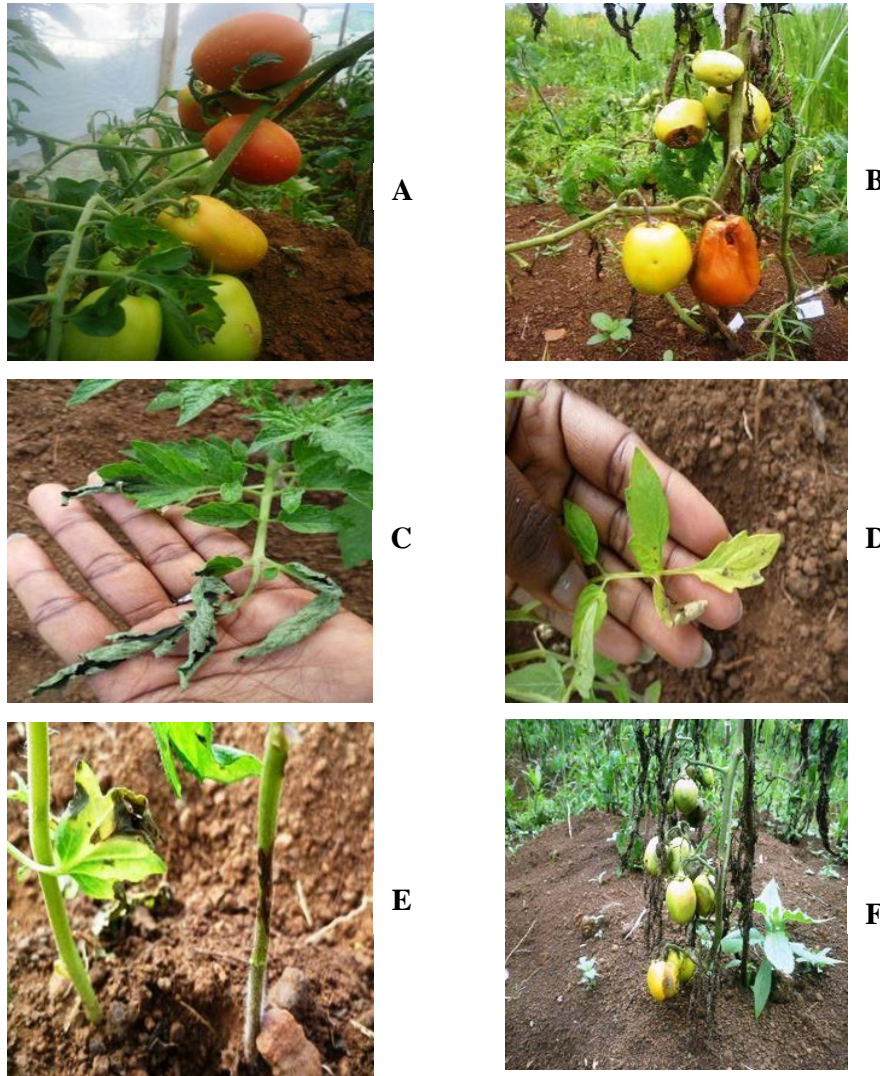


Fig. 6. Status of some disease symptoms compared to a healthy plant

Healthy plant and fruits (A); Diseased plant and fruits (B): Apical leaves shadding (C); leaves yellowish (D); Stem spot or necrosis (E); Total shadding of plant (F).

Table 4. Variation of main symptoms as influenced by treatments

Observed symptoms	Treatments					
	Tm-	Tm+	Tci	Tcii	Cpi	Cpii
Stem and foliar spots	++++	+++	++	++	+	+
shadding	+++++	+++	++	++	+	+
stunted	+++++	+++	+	+		
Spoilage at the bottom of fruits	+++++	++	+	+	+	+
Spots on fruits	+++++	++	++	++	+	+
Yellowish of leaves	++++	+++	++	++	+	+
Total spoilage of fruits	++++	++	+	+	+	+

[0-2] diseased plants or fruits/plant: +; [2-4] diseased plants or fruits/plant: ++; [4-6] diseased plants or fruits/plant: +++; [4-8] diseased plants or fruits/plant: ++++; [8-10] diseased plants or fruits/plant: +++++

All treatments presented all the symptoms, but symptoms such as shedding, black spots on fruits, yellowish of leaves, spoilage of fruits were less observed on Tci, Tcii, Cpi et Cpii treated plants than on Tm- and Tm+ plants. Stunted of plants was completely inhibited by Cpi and Cpii treatments (Table 4), confirming the findings of Elad and Shtienberg (1994) who postulated that the type of the compost does not directly affect the efficacy of the tea, and hence, both manure-based composts and other compost types can provide an efficient tea to control plant pathogens. A positive and significant correlation was found between the number of plant leaves and height ($r = 0.79$; $p < 0.0001$) on one hand, and the number of flowers ($r = 0.61$; $p < 0.0001$) on the other, implying that these parameters are closely related.

Conclusion

From this study, it is established that cow dung compost and compost tea are more effective and efficient in reducing diseases incidence on tomato plants than kitchen manure compost and compost tea. Hence, this is accomplished through stimulation of growth, reduction of disease symptoms, and suppression of foliar pathogens. They act by both their chemical composition and microorganisms and could therefore be used at appropriate proportions as biological fertilizers/pathogen antagonists. This result complies with those reported in a previous study on suppressive activity of compost and compost tea on *Solanum tuberosum* in the same agro-ecological zone. Since a biological control system requires a better knowledge of the inhibitive agent action mechanism, further researches will be undertaken to identify all the microorganisms involved thereof, and determine their suppressiveness mechanisms.

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