Evaluation of Neem Seed Extract for the Management of Early Blight (*Alternaria solani*) Disease of Tomato (*Solanum lycopersicum* L.)

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Summary

Early bight disease of tomato caused by the fungus, *Alternaria solani*, accounts for significant yield losses in tomato production annually. This research evaluated the use of aqueous neem seed extract for the management of early blight disease of tomato (*Solanum lycopersicon* L.) in a screenhouse experiment. Infected samples of tomato leaves and fruits were collected from an experimental plot at the University of Ibadan for isolation of associated fungi. Five kilogrammes of sterilized soil were filled into 20cm diameter experimental pots laid out in a completely randomized design with six treatments and three replications. A susceptible variety of tomato ('Cobra') was used as the test crop. Three concentrations, 50 g, 100 g, and 200 g per litre of neem seed extract were applied a week before and after inoculation.Synthetic mancozeb fungicide was used as positive control. The inoculum was quantified using the serial dilution method, and a concentration of 10^6 condidia/ml was applied on the plants. Pathogenicity, incidence and severity of isolates were determined following standard procedures. Data were collected on number of leaves, plant height (cm), stem diameter (cm), cumulative fruit weight (g) and total dry matter yield (g) per plant. Plants inoculated and sprayed with aqueous neem seed extract had the lowest disease incidence (40%) and severity score (1) of the early bight disease relative to other treatments. Plants inoculated and sprayed with aqueous neem seed extract also showed significant increase in yield and growth parameters and compared favourably with mancozeb synthetic fungicide. Neem concentrations used in this study reduced the incidence and severity of early blight disease to 33.3-45.1% and 20.7-29.4%, respectively relative to plants treated with mancozeb fungicide (12.8% and 10.6%, respectively). Although aqueous neem seed extract showed

potential in the management of early blight disease of tomato, further studies and field investigations should be carried out to explore the possible application of neem seed extract in the integrated management of the disease.

Keywords: Neem seed extract, Pathogenicity, *Alternaria solani,* Early blight, Conidia

TOMATO (*Solanum lycopercicum* L.) is a vegetable crop which belongs to the nightshade family, Solanaceae, and it is widely cultivated for its fruits throughout the humid and subtropics. Its production is severely affected by several diseases caused by fungi, bacteria, viruses and nematodes at all growing stages from seedling to maturity,resulting in considerable reduction in yield (36). Of these diseases, early blight caused by the necrotrophic fungus *Alternaria solani* (Ellis and Martin) Jones and Grout, is one of the most devastating foliar diseases occurring over a wide range of climatic conditions (15). The disease can lead to complete defoliationin areas with high relative humidity greater than 75% and temperature of between 24and29°C, and in semiarid climates where frequent and prolonged night dews occur (8, 28). Tomato plants are most susceptible at 8-10 weeks of growth.

Symptoms associated with the disease include collar rot or basal stem lesions at the seedling stage, extensive necrosis surrounded by yellow halo on leaves, stem lesions and fruit rot in the adult plant stage (11, 33).The leaf blight phase, commonly referred to as early blight, is the most important phase of the disease and can result in complete loss of the crop when incidence is severe (17). Collar rot due to early blight damage can cause seedling losses in the field from 20 to 40% and yield losses up to 79% (34). The disease occurs in major tomato-producing areas and symptoms develop more rapidly during periods when favourable environmental conditions alternate between humidity and drought(21, 35).

The main methods of controlling *Alternaria* leaf blight include preventing long periods of wetness on the leaf surface, sanitation, cultivation of resistant varieties and application of fungicides (19, 27). It has been reported that cultivation of resistant varieties is the ultimate control of this disease, but some tomato cultivars have been reported to show low level of genetic resistance to *Alternaria* leaf blight (13). Besides, farmers in pursuance of high yield are inclined to cultivate some varieties which may be less resistant to disease. Chemical control using synthetic fungicides is not a viable option either, due to

environmental concerns, pathogen resurgence and human safety. Hence, it has become imperative to explore alternative measures of disease control.

Organic pesticides are increasingly becoming popular in the management of plant diseases because their products have been found to be biodegradable, eco-friendly and safe for human health (5, 12, and 20). Due to its efficacy and minimum side effects, azadirachtin, a tetra terpenoid obtained from neem seeds, has emerged as a natural biopesticide and a viable alternative to synthetic pesticides (23, 29). Neem (*Azadirachta indica* A. Juss.) is an evergreen tree belonging to the family Meliaceae. It thrives well in tropical and sub-tropical regions, having unique attributes of fast growth and resistance to drought conditions.All parts of the tree including seeds, leaves, roots, bark, constitute a rich source of medicinal drugs (6, 31). The tree has been reported to possess insecticidal, antiseptic, antifungal, antibacterial and anti-malaria properties among several other uses (10, 25, 30). Neem contains a vast array of biologically active compounds that are chemically diverse and structurally variable with more than 140 compounds isolated from different parts of the tree (39). Quercetin and ß-sitosterol, were the first polyphenolic flavonoids purified from neem tree and were known to have antibacterial and antifungal properties (22). Also, neem is readily available in Nigeria and cheap unlike the synthetic pesticides. This study, therefore, evaluated the potential of neem seed extract in the management of early blight disease of tomato.

MATERIALS AND METHODS

Field sampling for early blight disease and isolation of associated fungi

Infected tomato leaf samples showing characteristic symptoms of early blight disease such as circular irregular black or brown spots on the older leaves of the plants including series of distinct dark concentric rings in the center of the spots were collected from an experimental plot at the roof top garden of the Department of Crop Protection and Environmental Biology (CPEB), University of Ibadan, southwest Nigeria using random sampling technique. Fifty diseased specimens were collected for isolation of infecting fungi; while seeds of a susceptible tomato variety, 'Cobra' were used for the experiment. Infected leaves were washed with sterile distilled water (SDW), surface-sterilized in 10% sodium hypochlorite for 1 minute, rinsed with three changes of SDW to remove surface contaminants and air-dried on

sterilized tissue paper. Plating of samples was done on potato dextrose agar (PDA) medium dispensed in 9 cm diameter Petri dishes and incubated at 28 ± 2^0C under alternating conditions of 12 - hour darkness and light. *Alternariasolani* was further sub - cultured on Czapeck agar to enhance sporulation, while other fungi sporulated on PDA. A litre of the medium consisted of Sucrose 30 g, Sodium nitrate 2g, Dipotassium phosphate1g, Magnesium sulphate 0.500 g, Potassium chloride 0.500g, Ferrous sulphate 0.010, and agar-agar 15g.

Identification of isolated fungi

The various fungal isolates from each of the samples were sub-cultured on PDA to obtain pure cultures for identification. The cultural features of each fungal isolate were carefully observed and recorded. Wet mounts of each isolate were prepared on a microscope slide and stained with lactophenol cotton blue. Themounts were then observed under a microscope and detailed structural features of the isolates were recorded. The features of the organisms were compared with those described in a standard manual of fungi following standard procedures (4, 9, 37). Further confirmation of isolates was made at the Mycological Herbarium in the Germplasm Health Unit of the International Institute of Tropical Agriculture (IITA), Ibadan.

Pathogenicity of isolates

The experiment for pathogenicity was laid out in a completely randomized design with three replications. The treatments consisted of three fungi isolated from the infected tomato samples: *Fusarium oxysporum*, *Colletotrichum* sp. and *Alternaria solani*. Seeds of a susceptible tomato variety, 'Cobra' were sown in sterilized soil and transplanted into experimental pots filled with 5 kg sterilized soil at three weeks after planting (WAP). At three weeks after transplanting, the seedlings were inoculated with a 10^6 conidial/ml of the test isolates; inoculum concentration was quantified using serial dilution and sprayed on the leaves using a hand sprayer. Control treatment was set up in a similar manner, except that sterile distilled water was used instead of inoculum. Symptom development was evaluated at 14 days after inoculation. Re-isolation and reinoculation was again done to establish Koch's postulates for pathogenicity.

Nursery bed preparation and experimental layout

Loamy soil for seedbed nursery and planting was obtained from the Crop Garden of the Department of Crop Protection and Environmental

Biology, University of Ibadan. The soil was sterilized at 170°C for 4 hours in an electrical sterilizer (Model SS-5-20, Philip Avent, USA) to eliminate microbial contamination. Sterilized soil was poured into a sterilized wooden tray measuring 90 cm × 40 cm which served as the nursery bed, and sufficiently watered before seeds of a susceptible tomato variety ('Cobra') were sown by drilling. Germinated tomato seedlings were transplanted after 3 weeks to experimental pots measuring to 20 cm in diameter and filled with 5kg of soil at one seedling per pot in a screenhouse. The experiment was laid out in a completely randomized design with six treatments and three replicates. The treatments were as follows:

Treatment 1: Plants were sprayed with sterile distilled water and served as control.

Treatment2: Plants were inoculated with the pathogen, but no fungicide or crude neem seed extract application

Treatment 3: Plants were inoculated with the pathogen and sprayed with 50g/L of crude neem seed extract.

Treatment 4: Plants were inoculated with the pathogen and sprayed with 100g/L of crude neem seed extract.

Treatment 5: Plants were inoculated with the pathogen and sprayed with 200g/L of crude neem seed extract.

Treatment 6: Plants were inoculated with the pathogen and sprayed with 0.5g/L of mancozeb fungicide.

Preparation of crude neem seed extract and inoculation of seedlings

Freshly harvested healthy neem seeds were air-dried at room temperature for two weeks and then, ground with a high-speed rotary blender (WPB80 Model 1122217,Warring, USA) into fine powder. The powder was dissolved in weights of 50, 100 and 200g per litre of sterile distilled water while 0.5g Mancozeb fungicide was dissolved per litre. The crude extract and fungicide were applied 2 times on the plant before and after inoculation using a hand sprayer. A 10-fold serial dilution method was used for inoculum preparation and a concentration of 10^6 conidia/mL was used for inoculation using a hand sprayer. Control treatment was sprayed with sterile distilled water. Data on number of leaves, stem

diameter, plant height, disease incidence and severity were collected weekly, while total fruit weight and biomass were collected at 12WAT. Number of leaves was determined by visual counting, plant height and stem diameter were measured using meter rule and vernier caliper (Model 830-104, Tresna, USA) respectively.

Determination of dry matter

The tomato plants were harvested from the middle of each plot by uprooting. The roots of the freshly harvested plants were washed with running tap water and plants from each plot were separately weighed to determine the total fresh weight. This was followed by partitioning of the component parts of each plant in order to determine shoot fresh weight and root fresh weight. The shoot was also partitioned into stem and leaves to determine fresh stem weight and fresh leaf weight. These were separately packed into paper bags. The samples were labelled and placed in an oven at 70^0C for 72 hours until constant weight was achieved. The samples were weighed using an electric Mettler balance Model PI210, USA to express the dry weight in grammes (14).

Determination of disease incidence and severity

Assessment of disease incidence and severity started at two weeks after transplanting (WAT). Disease incidence was determined as percentage of infected plants. This was calculated thus:

$$\text{Percentage disease incidence} = C/D \times 100$$

Where C = number of plants infected in a treatment

D = total number of plants in the treatment

Disease severity was rated on a scale of 1-5 following standard procedure (3).

1 = No symptom (0% leaf tissue infected)

2 = Mild symptom (<5% leaf tissue infected)

3 = Moderate symptom (>5%≤ 30% leaf tissue infected)

4 = Severe symptom (> 30% ≤ 50% leaf tissue infected)

5 = Very severe symptom (> 50% leaf tissue infected)

Thus, disease severity was calculated as:

$$\text{Disease severity index (\%)} = \frac{\text{Sum of disease ratings}}{\text{Total number of ratings x Maximum disease grade}} \times \frac{100}{1}$$

Data analysis

All data were subjected to analysis of variance (ANOVA) and means were separated using Duncan Multiple Range Test (DMRT). Least Significant Difference (LSD) was determined at 5% level of probability using SAS version 9.1 (38).

RESULTS

Pathogenicity, disease incidence and severity

Three fungal isolates: *Colletotrichum* sp., *Fusarium oxysporum* and *Alternaria solani* were associated with early blight disease symptoms, with only *A. solani* being pathogenic on re-inoculation following Kock's postulates. *Colletotrichum* sp., *Fusarium oxysporum* tested negative in the pathogenicity test and were therefore discarded.Disease assessment showed that the plants inoculated with *Alternaria solani*, but unsprayed with either neem seed extract or mancozeb fungicide had the significantly ($p<0.05$) highest incidence of 17.7% relative to other treatments and control with 0% incidence (Table 1). This trend continued progressively until 8WAT with all inoculated plants in that treatment, which showed early blight symptoms and recorded 85.4% incidence. Similarly, the same treatment exhibited the highest severity of disease symptoms of 78.7%. Tomato plants that were inoculated with the test pathogen and sprayed with mancozeb (T6) showed the least incidence and disease severity of 12.8% and 10.6%, respectively at 8WAT compared to all the other treatments. Mean incidence and severity were lowest at 4WAT but increased progressively reaching a maximum of 36.4 % and 26.7%, respectively.

Table 1: Effect of neem seed extract on percent incidence and severity of early blight disease symptoms

	2WAT		3WAT		4WAT		5WAT		6WAT	
Treatment	I*	S**	I	S	I	S	I	S	I	S
T1	0.0b	0.0b	0.0b	0.0b	0.0b	0.0b	0.0c	0.0c	0.00d	0.0c
T2	17.7a	14.8a	20.5a	37.7a	53.3a	64.8a	69.3a	73.2a	85.4a	78.7a
T3	3.8b	3.1b	8.8b	12.4b	17.2b	18.3b	40.4b	24.8b	45.1b	29.4b
T4	4.1b	2.8b	7.4b	10.7b	14.4b	14.1b	34.3b	19.6b	41.6b	20.9b
T5	2.7b	2.4b	5.5b	9.8b	15.8b	11.9b	32.2b	15.2b	33.3b	20.7b
T6	1.5b	2.4b	3.9b	3.1b	4.3b	5.7b	7.7c	8.2b	12.8c	10.6b
SD	6.4	5.3	7.0	13.3	18.8	23.2	24.8	25.9	29.6	27.4

*Incidence; **Severity; WAT= Weeks after transplanting; T1=Uninoculated control, T2 =Inoculated but unsprayed, T3=Inoculated and sprayed (50g/L extract), T4=Inoculated and sprayed (100g/L), T5=Inoculated and sprayed (200g/L), T6=Inoculated and sprayed Mancozeb fungicide (0.5g/L).

Means followed by same letter along a column are not significantly different at p = 0.05, using Duncan's Multiple Range Test (DMRT).

Effect of aqueous neem seed extract on number of leaves per tomato plant inoculated with *A. solani*

Tomato plants that were inoculated and sprayed with synthetic mancozeb fungicide produced the significantly ($p<0.05$) highest mean number of leaves (58.5) and 97.0 at 2 and 4WAT, respectively. This was closely followed by treatment sprayed with 50g/L of aqueous neem seed extract (53.1) but there was no significant difference between the two treatments ($p>0.05$) (Fig. 1). The application of 50g/L of the extract apparently encouraged production of significantly ($p<0.05$) highest number of leaves (115) at 6WAT relative to other treatments and control with no significant difference between both treatments. At 8 WAT, the plants inoculated and sprayed with 100g/L of aqueous neem seed extract (T4) produced the highest mean number of leaves (90.5) at 8WAT. This was followed by the plants inoculated and sprayed with 50g/L and 200g/L of aqueous neem seed extract with mean number of leaves of 90 and 89, respectively relative to the control. There were no significant differences ($p>0.05$) amongst the treatments.However, the plants that were inoculated and sprayed with aqueous neem seed extract and those that were inoculated with the pathogen but unsprayed differed significantly ($p<0.05$).

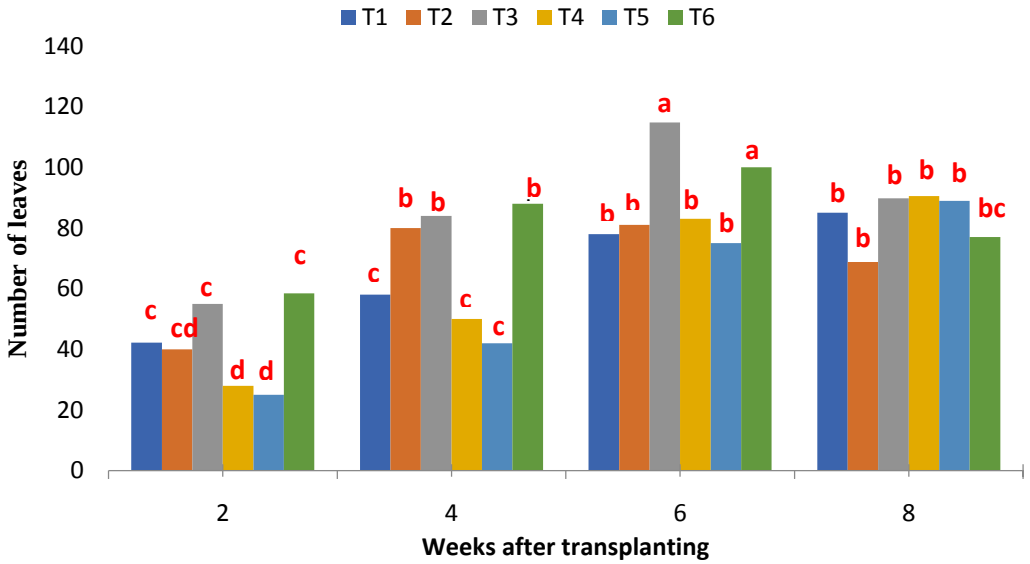

Figure 1: Effect of aqueous neem seed extract on number of tomato leaves inoculated with *A. solani*

T1= Control, T2 = Inoculated (unsprayed), T3= Inoculated and sprayed 50 g/L, T4= Inoculated and sprayed 100 g/L, T5= Inoculated and sprayed 200 g/L, T6= Mancozeb 0.5 g/L

Effect of aqueous neem seed extract on height of tomato plants inoculated with *A. solani*

The plants inoculated and sprayed with fungicide (mancozeb) were the tallest (27.6 cm) at 2 WAT. This was followed by those inoculated and unsprayed with inoculum and those that were inoculated and sprayed with 50 g/L of the extract with plant heights of 26.9 cm and 26.2 cm, respectively (Fig. 2). However, there were no significant differences ($p>0.05$) among the treatments at 2 WAT. Similarly, tomato plants that were sprayed with mancozeb recorded the highest plant height at 4WAT. This was again followed by the plants inoculated and unsprayed (T2) with a mean height of 42.5 cm. There was no significant difference among tomato plants that were inoculated but unsprayed with either neem seed extract or mancozeb fungicide at 7 and 8WAT.

Effect of aqueous neem seed extract on stem diameter of tomato plants inoculated with *A. solani*

Tomato plants that were inoculated and sprayed with mancozeb fungicide had significantly ($p < 0.05$) highest

mean stem diameter of 0.39 cm followed by plants inoculated and unsprayed (0.38 cm) at 2 WAT (Fig. 3). The lowest mean stem diameter of 0.28 cm was obtained from the plants that were inoculated with the pathogen and sprayed with 200 g/L of the extract. There were however, no significant ($p > 0.05$) differences among the five treatments. The plants inoculated and sprayed with mancozeb had the highest mean stem diameter of 0.62 cm and was significantly different from all other treatments at 4WAT. On the contrary, plants that were inoculated and sprayed with 200g/L of aqueous neem seed extract had the lowest mean stem diameter of 0.28 cm and were significantly different from other treatments at the same period. Similarly, mean stem diameter at 6 and 8 WAT was 0.61 and 0.60 cm, respectively among plants that were inoculated with the test pathogen and sprayed with mancozeb, which differed significantly from the other treatments

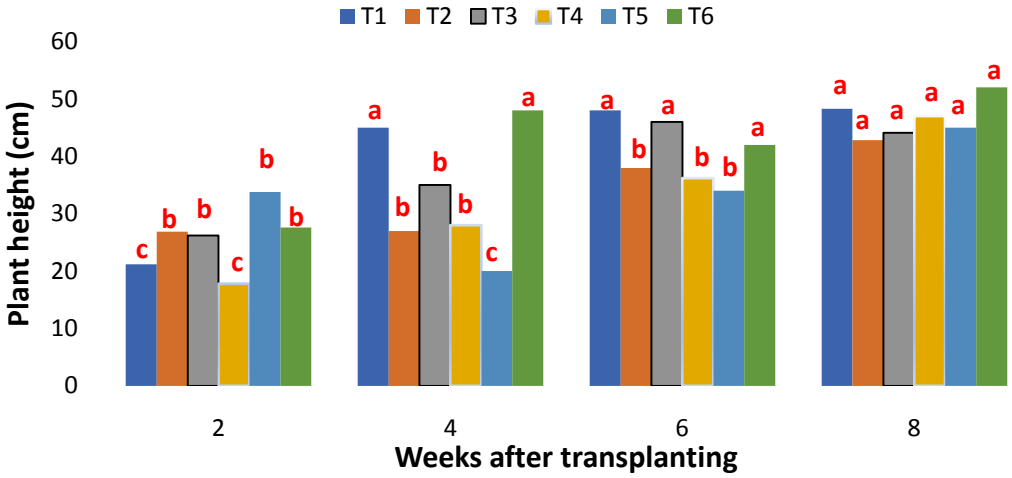

Figure 2: Effect of aqueous neem seed extract on height of tomato plants inoculated with *A. solani*

T1= Control, T2= Inoculated (unsprayed), T3= Inoculated and sprayed 50 g/L, T4= Inoculated and sprayed 100 g/L, T5= Inoculated and sprayed 200 g/L, T6= Mancozeb 0.5 g/L

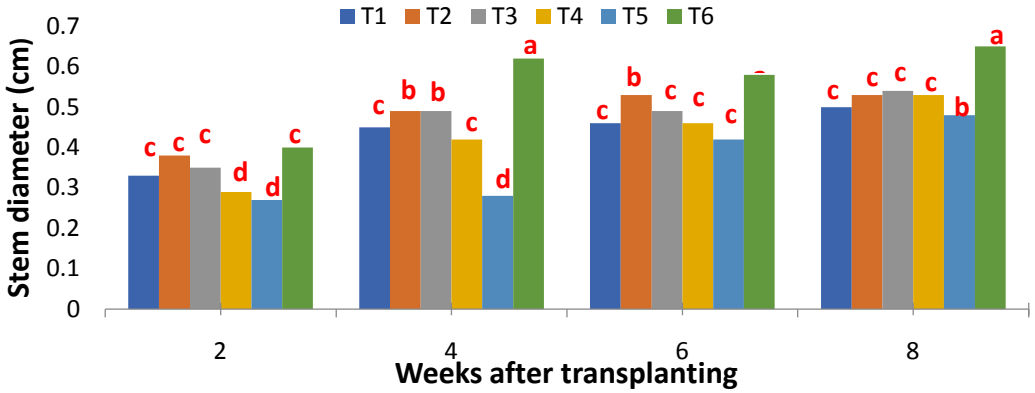

Figure 3: Effect of aqueous neem seed extract on stem diameter of tomato plant inoculated with *A. solani*

T1= Control, T2= Inoculated (unsprayed), T3= Inoculated and sprayed 50 g/L, T4= Inoculated and sprayed 100 g/L, T5= Inoculated and sprayed 200 g/L, T6= Mancozeb 0.5 g/L

Effect of aqueous neem seed extract on cumulative fruit yield of tomato inoculated with *A. solani*

Tomato plants that were inoculated and sprayed with mancozeb (T6) produced significantly ($p<0.05$) highest cumulative fruit yield (119.9 g) (Fig. 4). The lowest cumulative fruit yield of 34.5 g was recorded in treatment 2 which consisted of plants that were inoculated with *Alternaria solani* but unsprayed with either neem seed extract or mancozeb fungicide and was significantly different ($P<0.05$) from other treatments.

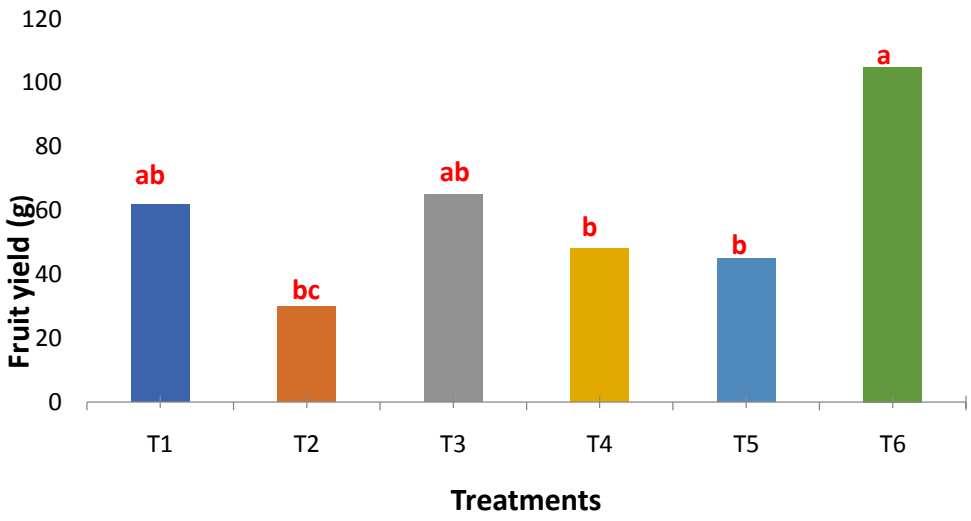

Figure 4: Effect of aqueous neem seed extract on cumulative fruit yield of tomato plants inoculated with *A. solani*

T1= Control, T2= Inoculated (unsprayed), T3= Inoculated and sprayed 50 g/L, T4= Inoculated and sprayed 100 g/L, T5= Inoculated and sprayed 200 g/L, T6= Mancozeb 0.5 g/L

Effect of aqueous neem seed extract on total dry matter of tomato plants inoculated with *A. solani*

The tomato plants inoculated and sprayed with 50g/L of aqueous neem seed extract had the highest total dry matter yield of 11.1 g (Fig. 5) followed by plants that were inoculated and sprayed with mancozeb (10.1 g). Control plants that were sprayed with sterile distilled water produced the lowest dry matter yield of 6.96 g and differed significantly from other treatments.

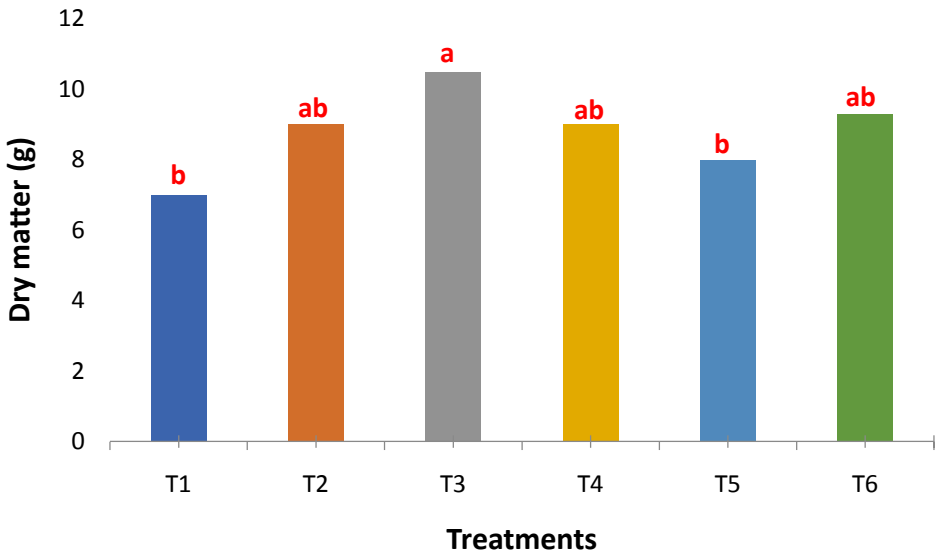

Figure 5: Effect of aqueous neem seed extract on total dry matter of tomato plants inoculated with *A. solani*

T1= Control, T2= Inoculated (unsprayed), T3= Inoculated & sprayed 50 g/L, T4= Inoculated and sprayed 100 g/L, T5= Inoculated and sprayed 200 g/L, T6= Mancozeb 0.5 g/L

DISCUSSION

Earlyleaf blight disease of tomato caused by the fungus, *Alternaria solani*, is a devastating disease of tomato which causes significant reduction in yield, quality and market value of the crop. It is an important disease which is prevalent in the tropical and sub-tropical regions of the world. The early blight pathogen can cause disease on all parts including the leaves, stems and fruits of the plant which results in severe damage during all stages of plant development (1). However, blight symptoms were more predominant on the tomato leaves because they were the site of application of inoculums. Although three fungal isolates, *Fusarium oxysporum*, *Colletotrichum* sp. and *Alternaria solani* were associated with early blight disease symptoms in this study, only *A. solani* tested positive in the pathogenicity study. Several authors also give credence to *A. solani* as the causal organism of early blight disease of tomato (13, 15, 21). The highest incidence and severity of early blight disease symptoms were

observed on the leaves of treatment inoculated with *A. solani* but unsprayed with neither neem seed extract nor synthetic mancozeb fungicide. The non-application of pesticides to the treatment was probably responsible for the high rate of infection since the activity of the blight-inducing pathogen was not inhibited. On the contrary, lower incidence and severity rates were observed in plants inoculated but sprayed with neem seed extract and mancozeb, which implied that the inhibitory action of the extracts reduced disease incidence and severity in the test plants. Previous authors had reported high disease incidence and severity of early blight among susceptible tomato varieties inoculated with *A. solani* (24, 27, 30). Plants sprayed with 50-200 g/L of neem seed extract had significantly lower disease incidence relative to the treatments that were inoculated but unsprayed with either the botanical or synthetic fungicide. This indicated that the extract competed favourably and even surpassed the efficacy of the synthetic fungicide. The potential of neem extract in the management of several pathogens and diseases had been articulated by previous authors. Hassanein *et al*. (16) conducted a research on the efficacy of neem leaves against blight and wilt inducing pathogens of tomato and found the botanical to have considerably reduced the incidence and severity of both diseases. Similarly, Bokhari *et al.* (7) reported a significant reduction in sclerotia formation and disease incidence in the evaluation of the efficacy of neem extract for the control of *Rhizoctonia solani* in infected soils. In another study, Adepoju *et al.* (2) found inhibitory effect of neem oil extract against four pathogens, *Fusarium* sp. *Rhizopus* sp. *Curvularia* sp. and *Aspergillus* sp. The significant reduction of disease incidence and severity among inoculated tomato plants, gives credence to the postulation that natural plant products are important sources of new agrochemicals for the control of plant diseases and the use of these environmentally safe methods in sustainable agriculture calls for reduction in the use of synthetic chemical fungicides (18, 32).

The application of aqueous neem seed extract significantly increased vegetative growth of the test plants, especially in the number of leaves per plant relative to plants that were sprayed with mancozeb. This implies that the neem seed extract in this study apparently enhanced vegetative growth better than fungicidal treatment with mancozeb. This result is consistent with previous findings of Nahak and Sahu (27) that evaluated the effect of neem leaf extract on

growth performance of tomato and reported a positive correlation between the application of the extract and the performance of the crop under field conditions. Similarly, Rajput *et al*. (31) reported the efficacy of neem products in the control of damping-off disease of shisham seedlings caused by *Fusarium solani*, and the enhancement of the overall growth of the test plants. Also, the plants that were inoculated with *A. solani* and sprayed with 50 g/L aqueous neem seed extract had significantly higher yields relative to those plants that were inoculated with the pathogen but unsprayed as well as other treatments. This showed that the application of neem seed extract did not only reduce the incidence of early blight disease, but also increased ultimate yield of tomato fruits. Conversely, the poor yield of treatments that were inoculated and unsprayed could be attributed to the unimpeded activity of the blight pathogen which was neither sprayed with the test extract nor the synthetic fungicide. Sale *et al*. (37) also conducted a research on the effect of neem extract preparations and nutrient sources on field performance of okra plants and reported impressive growth performance and yield from plants that were treated with the botanical. Moyin-Jesu (26) similarly established significant effect of modified neem leaf and wood extracts on disease reduction and soil improvement for the enhancement of growth and yield of water melon.

Although the synthetic mancozeb fungicide used in this study considerably reduced the incidence and severity of early blight disease, it could not completely eradicate the disease. Another deficiency is that the use of this synthetic fungicide in the management of the early blight disease of tomato is fraught with problems of environmental degradation, pathogen resurgence, elimination of natural enemies and other beneficial microbes in the ecosystem that make this option untenable. Neem seed extract compared favourably with mancozeb and significantly influenced vegetative growth and yield, with a concomitant reduction of disease incidence and severity on tomato plants evaluated. It is, therefore, strongly recommended in the management of early blight disease especially by virtue of being readily available, cheap, drought tolerant and eco-friendly with mininmal residual effect. More importantly, neem seed extract at lower concentration of 50 g/L enhanced better yields of the test plants than those obtained at higher concentrations of application, which makes its usage largely economical.

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