AGRICULTURAL AND BIOLOGICAL SCIENCES

# NEMATICIDAL POTENTIAL OF EXTRACTS OF NEEM (*Azadirachta indica*) AND LEMON GRASS (*Cymbopogon citratus*) ON ROOT-KNOT NEMATODES (*Meloidogyne* spp) INFECTING SWEET POTATO.

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**ABSTRACT**
The nematicidal potential of extracts of *Azadirachta indica* (neem) leaves and *Cymbopogon citratus* (lemon grass) on root-knot nematodes infecting sweet potato was evaluated between the months of June and November, 2012 at the Botanical Nursery of the University of Jos. The root-knot nematodes were obtained from galled roots of tomato and potato in Jos farms. A total of forty (40) sweet potato cultivar TIS 87/0087 were raised in steam-sterilized soil in clay pots and used for the study.Thirty (30) of the test plants were inoculated with 2250 juveniles of root-knot nematodes each while 10 were not inoculated serving as positive control. Ten of the inoculated plants were treated with extracts of neem leaves, 10 were treated with extracts of lemon grass while 10 were not treated, they served as negative control. Growth and yield parameters were then measured from 42 days after planting to 120 DAP using agronomic traits as a measure of growth and yield. Growth and yield parameters such as number of leaves, length of vines, number of tubers, weight of tubers etc were highest in the positive control, followed by sweet potato infected with nematodes and treated with lemon grass extract, then those treated with Neem leaves extract while those not treated with extract had the least growth and yield. Statistical analysis showed that the positive control produced significantly higher ($P< 0.05$) growth and yield parameters than all the other treatments. Infected plants treated with the extracts were also found to have significantly higher growth and yield parameters than the plants that were infected but not treated with extracts (negative control) at 0.05 level of probability. Number of galls was highest among the plants from the negative control, followed by plants treated with lemon grass extracts. The findings indicated that extracts of Lemon grass and Neem leaves improved growth and yield data of nematodes infected sweet potato and thus could be utilized in the control of root-knot nematodes in gardens.

**Key Words;** *Root-Knot Nematodes, Sweet Potato, Neem, Lemon Grass, Inoculate.*

## INTRODUCTION

The sweet potato (*Ipomoea batatas*) is a dicotyledonous plant that belongs to the family *Convolvulaceae* (Gills, 1988). It's large, starchy, sweet tasting, tuberous roots are an important root vegetable (Woolfe, 1992). The young leaves and shoots are sometimes eaten as greens. There are approximately 50 general and more than 1000 species of *Convolvulaceae*. *Ipomoea batatas* is the only genera of major importance, some others are used locally, but many are actually poisonous.

The genus, *Ipomoea* that contains the sweet potato also include several garden flowers called morning glories, though that term is not usually extended to *Ipomoea batatas*. Some cultivars of *Ipomoea batatas* are grown as ornamental plants; the name 'tuberous morning glory' may be used in a horticultural context (Purseglove, 1984). The plant does not tolerate frost (Onwueme, 1978). It grows best at an average temperature of $24^0$C ($75^0$F), abundant sunshine and warm nights. Annual rainfall of 750-1,000mm (30-39 in) is considered most suitable, with a minimum of 500mm (20 in) in the growing season. The crop is sensitive to drought at the tuber initiation stage (50-60 days after planting), and it is not tolerant to water logging, as it may cause tuber rots and reduce growth of storage roots if aeration is poor (Ahn, 1993). They grow well in many farming conditions and have few natural enemies, pesticides are rarely needed. Sweet potatoes are grown on a variety of soils, but well drained light to medium texture soils with pH range of 4.5 -7.0 are more favourable for the plant (Woolfe, 1992, and Ahn 1993).

Sweet potato is traditionally consumed in boiled form. The roots (tubers) are frequently boiled, fried or baked. They can also be processed to make starch and partial flour substitute (Purseglove 1984). Even the leaves of sweet potatoes plant have been shown to provide important antioxidant benefits and are included in soup in many cuisines (Chang, *et al.*, 2010).

Nematodes that attack plants are worms mostly microscopic in size, cylindrical in shape, tapering towards the head and tail.

Root-knot disease is caused by the root-knot nematodes *Meloidogyne* spp. Nematodes are microscopic warms that live in soil (Rasaki, 1981). Nematode feeds on plants roots, damaging and stunting them. The first evidence of a nematodes problem is poor growth of plants and poor colour of foliage (Jatala and Bridge, 1990). The damaged roots cannot supply sufficient water and nutrients to the above ground plant parts, and the plant is stunted or slowly dies. Symptoms are more pronounced during dry weather. Infested tubers are unsightly, but edible if peeled. The root-knot nematode cause small inconspicuous root swellings or galls to develop (Okechalu and Wonang, 2004).

The above ground symptoms exhibited by sweet potato plants infected by root-knot nematodes include stunting of plants, yellowing, wilting of plants, reduced yield and premature death of plants (IFAS, 2005). Small bumps or blisters on other varieties are important below-ground symptoms in sweet potato. There may be brown to black spots in the outer layers of flesh which are not evident unless the storage root is peeled.

Root-knot nematodes disease has been reported by some workers to be one of the major limitations of agriculture in the tropics (Adesiyan *et al.*, 1990). Recent approach to plant disease control de-emphasises the use of chemicals to control plant disease. Oyedunmade (2011) reported that extracts of plants such as lemon grass can be used as alternative to nematicides.Neem has also been reported to have nematicidal abilities (Kausik 2002) these, therefore, forms the background to this work which seeks to assess the nematicidal potential of extracts of Neem leaves and lemon grass on root knot nematodes infecting sweet potato.

## MATERIALS AND METHODS

The research work was carried out at the Botanical Garden of the University of Jos located on lat 08° 53'E and long 09° 57'N at an altitude of 1159m above sea level. Sweet potato cultivar TIS 87/0087 was used for the experiment. Extracts of Neem leaves (*Azadirachta indica*) and lemon grass (*Cymbopogon citratus*) collected from Jos and environs were tested on root-knot nematodes infecting sweet potato planted in clay pots.

Vines of sweet potato cultivar TIS 87/0087 were collected from National Root Crop Research Institute Sub-Station Maraba, Nasarawa State Nigeria on the 5th June, 2012. Lemon grass samples were collected from the University of Jos Staff Primary School. Neem leaves samples were collected from different locations within Jos, Plateau State

Samples of sweet potato plants together with the soil around their roots were collected from gardens behind the student village hostel of the University of Jos. A hand trowel was used to up- root the plants and a sharp knife to cut the root from top and carefully put inside black polythene bags. All the samples collected were taken to Plant Pathology Laboratory of the University of Jos for further processing.

Soil was collected from the garden in the Department of Plant Science and Technology, University of Jos, the soil was sterilised by heating it to a temperature of $80^0$C, then put in 40 clay pots and arranged into four rows, labelled as A, B, C and D, of 10 pots each, and were watered for 4 days before the

plants were introduced into it

Sweet potato vines collected were cut into 10cm as suggested by (Purseglove, 1984) and planted in the 40 pots. Lemon grass and Neem leaves were air dried and pulverised into powdered form and kept in separate containers. Afterwards, 50g of each extract was mixed in 1ml of water to obtain 50g/ml concentration of the extract.

Plant parasitic nematodes were extracted from the roots of sweet potato sourced from gardens around the Student's Village Hostel. The soil particles or sample collected from the adhering roots of sweet potato were carefully mixed to break up larger particles from the polythene bags then weighed out into 40g. The soil was then mixed with water for further processing.

The modified Baermann funnel technique as described by Southey (1970) was employed in extracting nematodes from the soil and root samples. This method is one of the fastest methods of recovering nematodes from roots and soil.

The nematode population was estimated thus; a drop (1 drop) of nematode extract viewed under microscope had 15 nematodes, therefore 10 drops contained 150 nematodes, 10 drops marked 1ml i.e.

1 drop = 15 nematodes
10 drops = 150 nematodes
10 drops = 1ml (which contained 150 nematodes)
15mls = 2250 nematodes

At three (3) Weeks after planting, sweet potato in plots A, B, and C were inoculated with 2250 Juveniles of root-knot nematodes each. This was done by pipetting 15mls of nematode suspension into a 5cm deep hole made near the root of each plant. After three weeks of inoculation of the plants with nematodes, 50g/ml extracts of lemon grass and Neem leave were applied to plots A and B respectively, and were covered with sand. Plot C had no extracts, they served as negative control while plot D served as positive control (i.e. was not inoculated with nematodes). Data on growth and yield parameters such as number of leaves per plant, length of vines per plant and number of galls per plant were all recorded at harvest. All data generated were subjected to statistical analysis using one-way analysis of variance.

## RESULTS AND DISCUSSION

The result of this investigation revealed that root knot nematode parasitize and reduce growth and yield of sweet potato plant with chlorosis on leaves of infected plants. The result also showed that the plant extracts used had nematicidal properties but to varying degree. Chlorosis of leaves was greatly noticed in inoculated but not treated plants. This result tallies with the observation of several workers (Sardenelli, 2010). He reported that root-knot nematode infection leads to physiological instability in plants that may be accompanied by symptoms such as chlorosis among others.

Mean number of leaves was highest (64.4) in plants grown in the control plot D (un-inoculated plants). This was followed by plants inoculated with nematodes and treated with lemon grass extract (47.6) then plants inoculated with nematodes and treated with neem leaves extract (47.4) while plants inoculated with nematodes but not treated with any extracts (the negative control) had the least mean number of leaves (30.2) (Table 1). The result also showed that the positive control plants had significantly higher mean number of leaves than all the other plants ($P<0.05$). Infected plants treated with neem and lemon grass did not differ from each other in mean number of leaves at 0.05 level of probability while infected plants not treated with any extracts had significantly lower mean number of leaves than all other treatments at 0.05 level of probability (Table 1).

Mean length of vines was highest (101.36cm) in plants grown in the positive control plots (i.e. un-inoculated) this was followed by plants inoculated with nematodes and treated with neem leaves extract (70.7) then plants inoculated with nematodes and treated with lemon grass extract (64.12) while plants inoculated with nematodes but not treated with any extract (negative control) had the least mean length of vine (33.04cm) (Table 1).

The result also showed that the un-inoculated plants (positive control) had significantly higher mean length of vine than all the other plants ($P<0.05$). Infected plants treated with neem extracts and infected plants treated with lemon grass did not differed from each other in mean length of vines at 0.05 level of probability while plants from the negative control had significantly lower mean length of vines as compared to all other plants at 0.05 level of probality (Table 1).

Mean number of tubers was highest in plants inoculated with nematodes and treated with lemon grass extracts (3.2) this was followed by plants grown in the positive control plots (un-inoculated) (3.1) while plants inoculated with nematodes but not treated with any extract (negative control) had the least mean number of tubers (1.5) (Table 1).

The result also showed that the plants inoculated with nematodes but not treated with any extract (the negative control) had significantly lower mean number of tubers than all the other treatments ($P<0.05$). Infected plants treated with neem did not differ from infected plants treated with lemon grass each at 0.05 level of probability (Table 1).

Mean weight of vines was highest (58g) in plants grown in the positive control plot (i.e. un-

inoculated) followed by plants inoculated with nematodes and treated with neem leave extract (45g) then plants inoculated with nematodes and treated with lemon grass extract (44.81g) while plants inoculated with nematodes but not treated with any extract (the negative control) had the least mean weight of vine (17.78g) (Table 2).

The result further showed that the positive control plants had significantly higher mean weight of vine than all the other plants ($P<0.05$). Also, the negative control had significantly lower mean weight of vine than all treatments at 0.05 level of probability while plants treated with lemon grass and neem leaves did not differ from each other in mean weight of vines (Table 2).

Mean weight of tubers was highest (81.4g) in plants grown in the positive control plots (un-inoculated) this was followed by plants inoculated with nematodes and treated with lemon grass extract (66.85g) then plants inoculated with nematodes and treated with neem leave extract (66.4g) while plants inoculated with nematodes but not treated (the negative control) had the least mean weight of tubers (17.1g) (Table 2). The observed better growth and yield of the positive control plants was not unexpected since they were not challenged with the nematodes.The two plants extracts i.e lemon grass and neem leaves improved growth and yield of nematode infected plants suggesting they had nematicidal properties on root knot nematodes.. This results tallies with the observation of Oyedunmade (2011) who conducted phytochemical analysis of lemon grass to determine the chemical compounds that had nematicidal activities. They reported that lemon grass controlled nematodes and consequently improved growth and yield of treated okra plants. Chopra and Hanson (1985) reported that lemon grass can be used as a nematicide. Also earlier reports by Sangwan *et al.,* (1985) showed that lemon grass has nematicidal properties. This may have accounted for the increased growth and yield in infected plants treated with lemon grass as observed in this study. Infected plants treated with neem leaves performed better than their untreated but infected controls. This observation is in agreement with the report by Kausik *et al., (*2002) who stated that neem leaves have nematicidal properties. Plants inoculated with nematodes and not treated with any extract showed greatest reduction in growth and yield parameters. This agrees with the report by Dropkin (1977) that nematodes cause loss of yield in crops. The observations in this study also tallies with the report of Adesiyan *et al.,* (1990) that a wide variety of crop plants are damaged by nematodes. These include roots and tuber crops such as cassava, yam, cocoyam, ginger and potato

Mean number of galls was highest (2.2) in plants inoculated with nematodes but not treated with any extract (the negative control) this was followed by plants inoculated with nematodes and treated with lemon grass extract (0.5) then plants inoculated with nematodes and treated with neem leaves extract (0.4) while plants grown in the control plots (uninoculated) had zero (0) mean number of galls (Table 2).

The result also showed that the plants inoculated with nematodes but not treated with any extract (the negative control) had significantly higher mean number of galls than all the other plants ($P<0.05$). All other treatments did not differ from each other in mean number of galls at 0.05 level of probability (Table 2). Galls on infected roots are characteristics symptoms of root-knot nematodes (Adesayan, *et al.,*1990) The plants showed greatest reduction in growth and yield with highest number of galls. The stunting of plants, chlorosis of leaves and poor yield are indicative of lack of physiological stability and it is often associated with reduced translocation, inadequate nutrients absorption and abnormal production of growth regulators as suggested by Wallace (1963). Reduction of yield can also be caused by pollen sterility induced by the nematodes as noted by Sasser and Carter (1975). These plants (negative control) showed highest root galling. Formation of galls on root is a distinctive feature of infection by root-knot nematodes (Bergeson, 1968).Uninoculated potato plants (positive control) were free from root galls. This may be because they were free from the root-knot nematodes.

Table 1: Mean Number of Leaves, Tubers and Length of Vine Per Sweet Potato Subjected to the varying treatments.

| Treatment | Mean No. of Leaves | Mean No. of Tubers | Mean Length Of Vine |
|---|---|---|---|
| A | 47.4 | 2.9 | 70.7 |
| B | 47.6 | 3.2 | 64.12 |
| C ( - Control) | 30.2 | 1.5 | 33.04 |
| D (+ Control) | 64.4 | 3.1 | 101.36 |
| LSD = | 16.44 | 0.83 | 10.87 |

Table 2: Mean Weight of Vine, Tubers and number of Galls per Sweet potato subjected to varying treatments.

| Treatment | Mean No. of Leaves | Mean No. of Tubers | Mean Length Of Vine |
|---|---|---|---|
| A | 45 | 66.4 | 0.4 |
| B | 44.81 | 66.85 | 0.5 |
| C ( - Control) | 17.78 | 17.1 | 2.2 |
| D(+Control) | 58 | 81.4 | 0 |
| LSD = | 11.68 | 11.06 | 0.65 |

Plate 1: Infected Sweet Potato Tubers

Plate 1: Sweet Potato Tubers showing signs of root-knot nematodes infection

## CONCLUSION

The result of this investigation has shown that neem leaves extract and lemon grass extract have nematicidal properties on root knot nematode infecting sweet potato. It is therefore suggested that leaves of these plants be utilized in the control of root-knot nematodes by farmers. This will reduce the dependence on pesticides which will go a long way in reducing cost of production as well as environmental effect of pesticides.

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