

NEMATICIDAL ACTIVITIES OF METHANOLIC LEAF EXTRACTS OF *AZADIRACHTA INDICA* A. JUSS AND *RICINUS COMMUNIS* L. ON THE ROOT-KNOT NEMATODE (*MELOIDOGYNE SPECIES*) OF POTATO (*SOLANUM TUBEROSUM* L.)

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ABSTRACT

The effects of methanolic leaf extracts of *Azadirachta indica* A. Juss. and *Ricinus communis* L. on root-knot nematode (RKN), *Meloidogyne* spp. infecting potato were investigated. In *in vitro* experiment, 88 juveniles of RKNs were exposed to different concentrations of the extracts (0.375 mg/ml, 0.75 mg/ml, 1.5 mg/ml and 3mg/ml) for 24, 48, 72 and 96 hours. Distilled water served as the control. The treatments were laid out in a completely randomized design (CRD) with four replicates. The nematodes were observed for mortality. Potato varieties Caruso and Yellow were used for the pot experiment. The potatoes were inoculated with 1,000 juveniles of RKN at 2WAP. The treatments included three concentrations each (0.75 mg/ml, 1.5 mg/ml and 3 mg/ml) of Neem and Castor; Furadan (7.7 mg/ml); inoculated + un-amended and un-inoculated + un-amended. All treatments were replicated five times in a CRD. The *in vitro* experiment showed an increase in nematode mortality with increasing concentration and duration of exposure for all treatments. Neem caused higher nematode mortality than castor. Potato treated with Neem extracts had the best performance. Furadan was most effective in reducing galling in the potatoes. Variety Caruso performed better than variety Yellow. Alkaloids, carbohydrates and tannins were present in the extracts. Nematicidal activities of neem and castor leaves were established.

Key words: Root-knot nematodes, botanical extracts, nematicide, potato, phytochemicals

INTRODUCTION

Potato (*Solanum tuberosum* L.) is one of the most important food crops in the world following rice, wheat and maize (CIP, 2018). It is an important cash crop in many countries (Tolno *et al.*, 2016). It provides more food per unit area than any other major crop (Devaux *et al.*, 2014). In terms of the number of producer countries, potato is second only to maize with more than 125 countries involved in its production worldwide; this is because potato is consumed by over a billion people across the globe (CIP, 2018).

Potato production in Africa tripled between 1994 and 2011 from 8 to 24 million metric tonnes, largely due to increase in the cropping area. Half of this production comes from sub-Saharan Africa (CIP, 2018). Egypt leads the production chart with an output of 5, 029, 022 tons followed by Algeria (4, 782, 690 tons) and South Africa (2, 150, 844 tons) (FAOSTAT, 2016). Nigeria has the largest land area dedicated to potato production in Africa with about 75% of the crop cultivated on the Jos-Plateau, but it is the sixth largest potato producer on the continent and ranks 39th in the world (FAOSTAT, 2016).

A study carried out by the International Potato Centre (CIP) and its partners from 2013 to 2016 in ten sub-Saharan African countries, including Nigeria, showed that famers are getting only a fourth of what they could produce on the same piece of land (8 tons/ha instead of 24 tons/ha) and that the current level of production could be increased by 140% if identified problems were addressed. The identified limitations were inadequate quality seeds and pests/diseases (CIP, 2018).

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Plant parasitic nematodes have been identified as a limiting factor in potato production, leading to decreased yield, poor tuber quality and deformations, which make them unmarketable (Medina *et al.*, 2016). Of all the nematodes parasitising potato, root-knot nematodes (RKNs) are the most aggressive, damaging and economically important (Jones *et al.*, 2013). Root-knot nematodes make host plants vulnerable to infections caused by other disease agents (Ekpenyong *et al.*, 2016). Nematodes have been reported to cause an estimated loss of about \$ 157 billion annually to world agriculture (Singh *et al.*, 2015). Loss of agricultural produce can be salvaged if these parasites are successfully combated (Tibugari *et al.*, 2012). The use of nematicides is the most effective method of controlling nematodes, but these are hazardous to both man and the environment (Ojo, 2016). Pesticide resistance, environmental degradation, human health impacts, resource losses and agronomic concerns have triggered moves to discourage chemical control to avoid environmental pollution (Ekpenyong *et al.*, 2016). The addition of organic content to the soil has been found to enhance the activity of naturally occurring biological organisms that compete with nematodes in the soil as well as exert nematicidal activity (Khan *et al.*, 2017). Therefore, the use of plants with nematicidal properties as organic soil amendment or their incorporation in compost compost manure can be effective, cheap and safe alternative methods for nematode control (Ekpenyong *et al.*, 2016). The present study was, therefore, designed to determine the nematicidal activities of methanolic leaf extracts of *Azadirachta indica* A. Juss and *Ricinus communis* L on RKNs infecting potato in order to determine the phytochemical composition of methanolic leaf extracts of the test plants, determine their *in vitro* nematicidal properties at different concentrations and durations of exposure as well as their effects at different concentrations when utilised as organic amendment on the growth and yield of potato inoculated with 1,000 second-stage juveniles of RKN.

MATERIALS AND METHODS

Experimental Site

The experiments were carried out in the laboratory and the Botanical Nursery of the Department of Plant Science and Biotechnology, University of Jos, Jos, Nigeria.

Collection of Test Plants

Young and fresh leaves of *Ricinus communis* L. and *Azadirachta indica* A. JUSS were collected from Ukadam, Bauchi ring-road, Jos and Sabon Gari (Yilpo), Mangu areas of Plateau State. The plant collection was done in September, 2018. The collected plants were identified by a taxonomist in the Department of Plant Science and Biotechnology, University of Jos.

Preparation of Test Plants for Methanolic Extraction

The fresh leaves of the test plants were washed under running tap water and shed-dried on a laboratory bench at room temperature. The leaves, dried to constant weight, were separately pulverised with clean pestle and mortar and then sieved with a 0.05 mm mesh size sieve. The leaf powder of each plant was kept in Bamma bottles for a day before subsequent extraction and phytochemical analyses.

Methanolic Extraction of the Test Plants

The extraction was done using the cold maceration technique. Fifty (50) g of *A. indica* and *R. communis* leaf powder was added to 500 ml of 70% methanol in separate, well labelled 1000 ml conical flasks and placed in a laboratory shaker at 60 rpm for 24 hours in the laboratory. The resulting mixture was filtered with a muslin bag and re-filtered through a Whatman No 1 filter paper. In each case, the filtrates were retained and the residues discarded. The filtrates were then evaporated to dryness in crucibles, over a water bath, at 100 °C.

Phytochemical Analysis of the Test Plants

The phytochemical analysis for qualitative detection of flavonoids, tannins, alkaloids, saponins, phlobatannin, cardiac glycosides, steroids, terpenoids and anthraquinone were performed on the extracts of both plants as described by AOAC (2007).

Sourcing of Potato Seed Tubers

Egg-size seed tubers of Caruso and Yellow varieties of potato were procured from Ribetshak N. C. S. Multipurpose Cooperative association, a potato farmers' cooperative association and a certified potato seed tuber producer in Sabon Gari (Yilpo), Mangu LGA of Plateau State.

Sourcing of Nematode Inoculum

Heavily galled tomato and okra roots, symptomatic of root-knot nematode infection, were collected at random from various irrigation fields in Farin-Gada and Lamingo within Jos, Plateau State, for the isolation of root-knot nematodes.

Isolation of Root-Knot Nematodes

Root-knot nematodes from infected tomato and okra roots were isolated using the modified Baermann Funnel method of nematode extraction as described by Sato *et al.* (2009). Galls were excised from the infected roots, dissected transversely and placed in Petri dishes containing about 10 ml distilled water. Using sterile inoculating needles, the galls were then teased to release the root-knot nematodes from the plant tissue. The samples were subjected to the Baermann funnel technique for nematode extraction. The Baermann funnel is a regular laboratory funnel with a piece of rubber tube about 25-30 cm long attached to the stem of the funnel. The tube is in turn connected to a test tube tightly held together with the aid of a masking tape. The set-up was kept in an upright position using a table stand with small regular holes and was filled with distilled water to the brim of the funnel's stem. Cotton wool was placed in the funnel to assume the shape of the funnel so that the water slightly covered the wool before the teased root samples for extraction of nematodes were placed on the cotton wool and covered with water to prevent the sample from drying. The set-up was allowed to stand for 24 hours. The active nematode juveniles readily passed through the cotton wool down the funnel stem and were collected at the bottom of the test tube. Twenty sets of the set-up were used so as to obtain sufficient quantities of the inoculum. The test tube was then carefully removed and its content centrifuged at 2,000 rpm (revolutions per minute) for 5 minutes to concentrate the nematode juveniles at the bottom of the test tube. After 24 hours, the test tubes containing the live nematodes were detached from the set-up and taken to the laboratory for population estimation, *in vitro* study and inoculation in the pot experiment.

Estimation of Nematode Population

Nematode populations were estimated by counting the number of active juveniles in 1 ml of homogenised suspension of root-knot nematodes under a binocular research light microscope at x40 magnification. One millilitre of homogenized suspension contained an average of 22 nematode juveniles.

Preparation of Extract Concentration for *In Vitro* and Pot Experiments

Various concentrations of the methanolic leaf extract of the test plants were prepared as follows: 1.5 g of each extract was dissolved in 250 ml of distilled water to yield a stock solution of 6 mg/ml. Other concentrations were separately prepared from the stock solution of each extract by two-fold dilution. Finally, the following concentrations were obtained: 3 mg/ml, 1.5 mg/ml, 0.7 mg/ml and 0.375 mg/ml.

***In Vitro* Nematicidal Determination**

Extract preparation and nematode extraction were carried out concurrently to ensure that fresh extracts and freshly extracted nematode juveniles were used throughout the experiment. One millilitre of each concentration of extract was added to 4 mls of homogenised suspension of root-knot nematode suspension (88 juveniles); this was replicated three times for each concentration and examined at 24, 48, 72 and 96 hours for live and immobilized nematodes. The experiment was laid out using the Completely Randomized Design with 2 plant extracts, 4 concentrations of each extract, a control (Nematode suspension + distilled water without extract) and 4 replicates for each treatment. The number of immobilized nematodes was counted and expressed as percentage of mortality and recorded. The immobile nematodes were then transferred to distilled water after 96 hours to ascertain whether or not they regained mobility. The mortality was confirmed by touching the nematode juveniles with a fine needle. Nematodes that appeared stiff and straight and did not move when probed with the fine needle were counted as dead nematodes (Muhammad *et al.*, 2004).

Pot Experiment

A completely randomised design was used in the pot experiment. Two potato varieties (Caruso and Yellow), three concentrations each of methanolic leaf extracts of *A. indica* and *R. communis* (0.75 mg/ml, 1.5 mg/ml and 3 mg/ml) and three controls i.e. a nematicide (Furadan at an application rate of 4 kg/ha which was equivalent to 7.7 mg/ml based on the area of polyethylene bags used for this study), distilled water and an un-inoculated + amended control were used. All treatments were replicated five times to give a plant population of 90. The potato plants were raised in steam-sterilized loamy soil in polyethylene pots 35 cm wide.

The plants were inoculated with 1,000 second-stage larvae of *Meloidogyne* species at 2 weeks after planting (2 WAP). This was done by pipetting 44 mls of the nematode solution into holes made around the base of the potatoes. The holes were carefully covered with soil. The various treatments were administered at 4 WAP by drenching each pot with 500 ml of the appropriate treatment. Two days later, more soil was added to the base of the plants (earthing /hilling up) to encourage tuberization.

The plants were carefully uprooted after 70 days (i.e. 70 WAP) to determine shoot height, number of tubers per plant, fresh tuber weight and the number of galls per plant. The roots were rated for galling index on a 0-5 scale (Taylor and Sasser, 1978), where 0 = no gall/plant, 1 = 1-2 galls/plant, 2 = 3-9 galls/plant, 3 = 10-30 galls/plant, 4 = 31-100 galls/plant and 5 = > 100 galls/plant.

Data Analysis

All data collected were analysed using two-way Analysis of Variance (ANOVA) test at 5 % level of probability. Means were separated using the Least Significant Difference (LSD) test.

RESULTS

***In-Vitro* Test**

Effect of Treatments on Nematode Mortality

The result revealed that nematode mortality increased with increase in time of exposure and concentration of extracts (Table 1). The highest nematode percentage mortality (100%) was recorded for 3 mg/ml of *A. indica* at 96 hours of exposure while 0.375 mg/ml of *R. Communis* resulted in the least mortality at 96 hours. Also, more than 50% mortality was recorded for all treatments (except for the control and 0.375 mg/ml of *R. Communis*) at 96 hours (Table 2). Generally, the different concentrations of *A. indica* leaf extracts had higher percentage mortality than those of *R. Communis*. Statistical analysis revealed that mean mortality due to the various concentrations of the two extracts were significantly higher than that of the control at 5 % level of probability.

The result also showed that mean mortality was significantly ($p < 0.05$) affected by time of exposure. Nematodes exposed to the extracts for 96 hours had significantly ($p < 0.05$) higher mortality than those from the other durations of exposure (Tables 1 and 2).

Table 1: Mean mortality of nematodes treated with varying concentrations of test plant extracts and the control at varying exposure time

Treatment	Concentration	Mean Nematode Mortality				LSD
		Time of Exposure (Hours)				
		24	48	72	96	
<i>Azadirachta indica</i>	0.375 mg/ml	19.00	19.75	32.50	44.00	4.90
	0.75 mg/ml	22.75	37.00	52.75	63.25	
	1.5 mg/ml	42.50	60.00	67.25	76.00	
	3 mg/ml	43.50	70.00	84.50	88.00	
<i>Ricinus communis</i>	0.375 mg/ml	14.25	18.50	32.50	42.50	
	0.75 mg/ml	22.75	25.25	38.75	50.25	
	1.5 mg/ml	31.50	37.25	57.00	64.75	
	3 mg/ml	41.75	53.00	76.75	82.50	
Control	Distilled water	0.00	1.75	2.75	7.50	
LSD		7.35				

Values are means of four replicates. Pairs of means which differ by more than their LSD value are significantly different at 5 % level of probability.

Table 2: Percentage mean mortality of nematodes treated with varying concentrations of test plant extracts and the control at varying exposure time

Treatment	Concentration	Percentage Mean Nematode Mortality				LSD
		Time of Exposure (Hours)				
		24	48	72	96	
<i>Azadirachta indica</i>	0.375 mg/ml	21.59	22.44	36.93	50.00	5.57
	0.75 mg/ml	25.85	42.05	59.94	71.88	
	1.5 mg/ml	48.30	68.18	76.42	86.36	
	3 mg/ml	49.43	79.55	96.02	100.00	
<i>Ricinus communis</i>	0.375 mg/ml	18.40	21.02	36.93	48.30	
	0.75 mg/ml	25.85	28.69	44.03	57.10	
	1.5mg/ml	35.80	42.33	64.77	73.58	
	3mg/ml	47.44	68.44	87.22	93.75	
Control		0.00	1.99	3.13	8.52	
LSD		8.35				

Values are means of four replicates. Pairs of means that differ by more than their LSD value are significantly different at 5 % level of probability.

Phytochemical Screening

The phytochemical screening of the methanolic leaf extracts of the test plants revealed that *A. indica* contained alkaloids, carbohydrates and tannins while *R. communis* contained carbohydrates and tannins (Table 3).

Table 3: Phytochemical profile of methanolic leaf extracts of *Azadirachta indica* and *Ricinus communis*

S/N	Biochemical	Test Plants	
		<i>A. indica</i>	<i>R. communis</i>
1	Alkaloids	+	-
2	Carbohydrates	+	+
3	Flavonoids	-	-
4	Cardiac glycosides	-	-
5	Phenols	-	-
6	Saponins	-	-
7	Sterols	-	-
8	Tannins	+	+
9	Terpenes	-	-

Key: + = Present; - = Absent

Pot Experiment**(a) Effect of Treatments on Shoot Height**

The result revealed that shoot height of extract-treated potatoes was higher than the un-amended control. Generally, variety Caruso had higher shoot height than variety yellow. Also, potatoes treated with *A. indica* had higher shoot height than those of *R. communis*. Generally, plant height increased with increasing concentration of extracts. Statistical analysis showed that shoot height for all treatments was significantly higher than those of inoculated and un-amended controls (Table 4).

Table 4: Mean shoot height of potato varieties at harvest treated with varying concentrations of the test plants.

Treatment	Concentration (mg/ml)	Mean Shoot Height (cm)		LSD
		Caruso	Yellow	
<i>Azadirachta indica</i>	0.75	43.8	43.6	0.50
	1.5	43.2	44.2	
	3	43.6	43.4	
<i>Ricinus communis</i>	0.75	43.8	44.2	
	1.5	44.2	43.2	
	3	43.4	44.4	
Furadan	7.7	44.0	43.2	
Inoculated, un-amended	Distilled water	37.2	37.0	
Un-inoculated, un-amended	Distilled water	43.8	42.4	
LSD		1.05		

Pairs of means that differ by more than their LSD value are significantly different at 5 % level of probability
Values are means five replicates

(b) Effect of Treatments on Number of Leaves

The result showed that potatoes grown in soils amended with the plant extracts had a higher number of leaves than the control. The highest number of leaves was recorded in pots amended with extracts of *A. indica* for both varieties of potato, followed by *R. communis*-amended pots, then un-inoculated and the untreated control, Furadan-treated

pots and the inoculated but untreated control in that order. Also, the number of leaves per plant stand increased with increasing concentration of the plant extracts. Generally, Caruso variety of potato had higher number of leaves than variety Yellow. Statistical analysis revealed that the observed differences in means were significant at 5 % level of probability (Table 5).

Table 5: Mean number of leaves plant of potato varieties treated with varying concentrations of the test plants at harvest

Treatment	Concentration (mg/ml)	Mean Number of leaves per plant		LSD
		Caruso	Yellow	
<i>Azardichta indica</i>	0.75	34.6	32.4	1.09
	1.5	36.2	36.0	
	3	38.0	36.0	
<i>Ricinus communis</i>	0.75	34.4	29.6	
	1.5	35.2	31.6	
	3	37.8	36.4	
Furadan	7.7	33.6	34.0	
Inoculated, un-amended	Distilled water	30.8	29.4	
Un-inoculated, un-amended	Distilled water	35.8	31.6	
LSD		2.31		

Pairs of means that differ by more than their LSD value are significantly different at 5 % level of probability. Values are means of five replicates

(c) Effect of Treatments on Number of Tubers

Number of tubers generally increased with increasing concentration of the plant extracts. Statistical analysis revealed that pots treated with extracts of *A. indica* had a significantly higher ($p < 0.05$) number of tubers than *R. communis*. There was no significant difference ($p < 0.05$) between pots treated with Furadan and the other two controls (inoculated + un-amended and un-inoculated + un-amended). There was a significant difference between the two varieties, with variety Caruso having a higher number of tubers than variety Yellow (Table 6).

Table 6: Mean number of tubers of the two potato varieties treated with varying concentrations of the test plant extracts

Treatment	Concentration	Mean Number of Tubers Per Stand		LSD
		Caruso	Yellow	
<i>Azadirachta indica</i>	0.75 mg/ml	9.6	7	1.3
	1.5 mg/ml	11.8	11.6	
	3 mg/ml	12.4	11.2	
<i>Ricinus communis</i>	0.75 mg/ml	5.6	7.2	
	1.5 mg/ml	9.5	6.2	
	3 mg/ml	12	6.6	
Furadan	7.7mg/ml	6.2	5.8	
Inoculated and un-amended	Distilled water	5.0	4.6	
Un-inoculated and un-amended	Distilled water	8.6	5.8	
LSD		2.8		

Values are means of five replicates. Pairs of means that differ from each other by more than their LSD value are significantly different at 5 % level of probability.

(d) Effect of Treatments on Fresh Tuber Weight

Tuber weight across the two varieties increased with increasing concentration of the plant extracts. Statistical analysis revealed that pots treated with extracts of *A. indica* had the highest tuber weight followed by *R. Communis*, un-inoculated + un-amended treatment and Furadan in that order. The least tuber weight was recorded in inoculated + un-amended pots. Statistical analysis also revealed that there was a significant difference between tuber weights of variety Caruso and variety Yellow with variety Caruso having higher tuber weight (Table 7).

Table 7: Mean fresh tuber weight of potato varieties treated with varying concentrations of test plant extracts

Treatment	Concentration (mg/ml)	Mean Fresh Tuber Weight (g)		LSD
		Caruso	Yellow	
<i>Azadirachta indica</i>	0.75	69.00	63.20	2.59
	1.50	77.00	75.20	
	3.00	80.60	73.20	
<i>Ricinus communis</i>	0.75	62.80	58.20	
	1.50	68.60	59.80	
	3.00	77.80	63.60	
Furadan	7.70	61.80	61.20	
Inoculated, un-amended	Distilled water	56.20	52.40	
Un-inoculated, Un-amended	Distilled water	63.60	60.50	
LSD		5.48		

Values are means of five replicates. Pairs of means that differ by more than their LSD value are significantly different at 5 % level of probability.

(e) Effect of Treatment on Gallings and Gall Index of Potato Roots

The result of the mean number of galls per plant revealed that the inoculated and untreated plants had the highest number of galls. Among amended plants, the number of galls decreased with increase in concentration. Generally, potatoes treated with varying concentrations of *A. indica* had lower number of galls compared to potatoes treated with *R. communis*. Variety Yellow was found to have higher number of galls than variety Caruso. Plants treated with Furadan had the least number of galls (Table 8). Statistical analysis showed that the observed variations between means across all treatments and the two varieties of potato were significant at 5 % level of probability (Table 8).

Table 8: Mean number of galls/plant of the potato varieties treated with varying concentrations of the plant extracts and the controls

Treatment	Concentration	Mean Number of Galls/Plant		LSD
		Caruso	Yellow	
<i>Azadirachta indica</i>	0.75 mg/ml	29.40	30.80	1.63
	1.5 mg/ml	25.60	27.00	
	3 mg/ml	21.80	22.80	
<i>Ricinus communis</i>	0.75 mg/ml	34.80	36.00	
	1.5 mg/ml	28.80	36.40	
	3 mg/ml	25.80	28.60	
Furadan	7.7 mg/ml	9.00	6.80	
Inoculated and un-amended	Distilled water	58.20	59.40	
Un-inoculated and un-amended	Distilled water	0.00	0.00	
LSD		3.45		

Values are means of five replicates. Pairs of means that differ from each other by more than their LSD value are significantly different at 5 % level of probability

Resistance rating showed variation in the plants' response to the varying treatments. Plants treated with Furadan were moderately resistant while plants treated with varying concentrations of the *A. indica* were moderately susceptible, except variety Yellow at 0.75 mg/ml concentration which was susceptible. Plants treated with *R. communis*, 3 mg/ml concentration and variety Yellow at a concentration of 1.5 mg/ml resulted in a resistance rating of moderately susceptible. All others were susceptible. The inoculated and un-amended plants were also susceptible (Table 9).

Table 9: Mean number of galls, galling index and resistance rating of potato varieties treated with varying concentrations of the plant extracts and the controls.

Variety	Treatment	Concentration (mg/ml)	Number of Galls	Gall Index	Resistance Rating
Caruso	<i>A. indica</i>	0.75	29.40	3	Moderately susceptible
		1.5	25.60	3	Moderately susceptible
		3	21.80	3	Moderately susceptible
	<i>R. communis</i>	0.75	34.80	4	Susceptible
		1.5	28.80	3	Moderately susceptible
		3	25.80	3	Moderately susceptible
	Furadan	7.7	9.00	2	Moderately resistant
	Inoculated & un-amended		58.20	4	Susceptible
	Un-inoculated & un-amended		0.00	0	Immune
Yellow	<i>A. indica</i>	0.75	30.8	4	Susceptible
		1.5	27.00	3	Moderately susceptible
		3	22.80	3	Moderately susceptible
	<i>R. communis</i>	0.75	36.00	4	Susceptible
		1.5	36.40	4	Susceptible
		3	28.60	3	Moderately susceptible
	Furadan	7.7	6.80	2	Moderately resistant
	Inoculated & un-amended		59.40	4	Susceptible
	Un-inoculated & un-amended		0.00	0	Immune

Gall index on 0-5 scale (Taylor and Sasser, 1978), where 0 = no gall/plant, 1 = 1-2 galls/plant, 2 = 3-9 galls/plant, 3 = 10-30 galls/plant, 4 = 31-100 galls/plant; and 5 = > 100 galls/plant. Values are means of five replicates

DISCUSSION

The phytochemical screening revealed that the leaf extracts of *Ricinus communis* L. and *Azadirachta indica* A. JUSS contained some bioactive constituents such as alkaloids, carbohydrates and tannins. It has been reported that bioactive compounds such as alkaloid, tannin, saponin, flavonoid, steroid, among others, have nematocidal properties (Chitwood, 2002). This may account for the nematocidal activities of the leaf extracts in the present study.

The nematocidal activities of the leaf extracts increased with concentration and time of exposure *in vitro*. This agrees with the report of Okechalu *et al.* (2020), who reported similar findings in the leaf extracts of *Commelina benghalensis* and *Bidens pilosa in vitro*. The observed mortality in the control treatment may have been as a result of starvation.

Growth and yield of potato treated with leaf extracts of *A. indica* and *R. communis* were higher than those of the control. The results indicated that *A. indica* and *R. communis* leaf extracts inhibited RKN invasion and lessened pest activity, resulting in the improved growth and yield. These findings agree with the reports of Khan *et al.* (2019) who observed that nematode infected crops treated with plant extracts had improved performance compared with their untreated counterparts. Potato plants that were inoculated and un-amended performed poorly, suggesting that the un-amended control plants were readily attacked by RKN as they exhibited the highest number of galls and the least growth and yield data. This finding is consistent with the report of Khan *et al.* (2019).

The extract-treated potatoes had root galling indices that were lower than the un-amended ones, as also reported by Hayat *et al.* (2012), that the use of parts of neem and neem products are very effective in combating root-knot nematode infections of cultivated crops. The decrease in gall formation in extract-treated pots may be attributed to the phytochemical composition of *A. indica* and *R. communis* which may be deleterious to RKN as suggested by Chitwood (2002). Although *A. indica* and *R. communis* extracts were less suppressing on gall formation than Furadan, their growth and yield data were significantly higher than Furadan and the other controls. This might be due to the nutrients added to the soil, which caused the plants to perform well in spite of the inoculation with nematodes.

CONCLUSION

The two soil amendments used in this study were effective in reducing the infection of potato by *Meloidogyne* species. However, *A. indica* was more effective in preventing the fecundity of root-knot nematodes and improving the yield and growth performance of potato. The control of plant-parasitic nematodes using Furadan was most effective in reducing the galling and gall indices of Caruso and Yellow varieties of potato. It is, therefore, concluded that Caruso variety of potato performed better than variety Yellow in terms of resistance to root-knot nematodes and growth and yield performance.

It is recommended that Caruso variety of potato be cultivated in nematode-infested soils since it demonstrated better resistance to root-knot nematodes than Yellow variety in this study. It is also recommended that nematode-infected plants be treated with extracts of *A. indica* and *R. communis* to improve crop yield and suppress nematode infestation below the threshold of economic loss.

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REFERENCES

- Association of Official Analytical Chemists (A. O. A. C.) (2007). *Official Methods of Analysis*, 18th Edition, Wilson Boulevard Publishers: Arlington, Virginia, USA.
- Chitwood, D. (2002). Phytochemical-Based Strategies for Nematode Control. *Annual Review of Phytopathology*, 40:221–249.
- CIP (2018). Potato Production in Africa can be Increased by 140%: find out how. https://cipotato.org/press_room/blogs/potato-production-in-africa-can-be-increased-by-140%-find-out-why/. Accessed July 11, 2018.
- Devaux, A., Kromann, P. and Ortiz, O. (2014). Potatoes for sustainable global food security. *Potato Research*, 5: 185–199.
- Ekpenyong, E.P., Ononuju, C.C., Orikara, C.C. and Ikwunagu, E. A. (2016). Nematicidal Potential of three Plant Materials Against Root-Knot Nematode (*Meloidogyne incognita*) Infestation on Tomato (*Solanum lycopersicum*). *International Journal of Life Sciences*, 4 (1):22-28.
- FAOSTAT (2016). Potato Production in 2016. Available from: <http://www.fao.org/faostat/en/#data/QC>. Retrieved October 20th, 2018.
- Hayat, A., Javed, N., Khan, S. A., Gondal, A. S. and Khan, H. U. (2012). Effect of Organic Amendments on Nematode Gallling Index and Egg Masses Production in Potato inoculated with Root-Knot Nematodes. *Pakistan Journal of Phytopathology*, 24(1): 1-5.
- Jones, J.T., Haegeman, A., Danchin, E.G., Gauer, H.S., Helder, J., Jones, M.G., Kikuch, T., ManzanillaLopes, R., Palomares-Rius, J.E., Wesemael, W.M. and Perry, R. N. (2013). Top 10 Plant-Parasitic Nematodes. *Molecular Plant Pathology*, 14: 946-961.
- Khan, R., Hussain, S., Khan, A., Ullah, S., Rashid, M. and Siddique, I. (2019). Phytochemical management of root-knot nematodes (*Meloidogyne incognita*) by *Artemisia* spp in tomato. *Brazilian Journal of Biology*, 10:590 – 600.
- Khan, A., Asif, M., Tariq, M. and Rehman, B. (2017). Phytochemical investigation of Nematostatic and Nematicidal potential of weed extracts against *Meloidogyne incognita* *in vitro*. *Asian Journal of Biological Sciences*, 10 (2):38- 46.
- Medina, I.L., Gomes, C.B., Correa, V.R., Mattos, V.S., Castagnone-Sereno, P. and Carneiro, R.M.D.G. (2016). Genetic Diversity of *Meloidogyne* spp. Parasitizing Potato in Brazil and Aggressiveness of *M. Javanica* Populations on Susceptible Cultivars. *Nematology*, 1:1-12.

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- Muhammad, S.A., Tariq, M. and Riaz, A. (2004). Some studies on the control of Citrus Nematodes (*Tylenchulus semipenetrans*) by Leaf Extracts of Three Plants and Their Effects on Plant Growth Variables. *Asian Journal of Plant Sciences*, 3(5): 544-548.
- Ojo, J. K. (2016). Pesticides use and health in Nigeria. *Ife Journal of Science*, 18 (4):1-11.
- Okechalu, O. B., Oyeyinka, J. and Danahap, L. S. (2020). *In Vitro* Evaluation of the Nematicidal Potential of Leaf Extracts of *Commenlina benghalensis* and *Bidens pilosa* on Root-Knot Nematode (*Meloidogyne* Spp). *Journal of Natural Sciences Research*, 10 (4): 32-41. www.iiste.org.
- Sato, E., Min, Y. Y., Toyota, K. and Takada, A. (2009). Relationships between the damage to radish caused by the root-lesion nematode *Pratylenchus penetrans*, its density prior to cultivation and the soil nematode community structure evaluated by polymerase chain reaction-denaturing gradient gel electrophoresis. *Soil Science and Plant Nutrition*, 55(4): 478-484.
- Singh, S., Singh. B. and Singh, A.P. (2015). Nematodes: A threat to sustainability of Agriculture. *Procedia Environmental Sciences*, 29: 215-216.
- Taylor, A. L. and Sasser, J.N. (1978). “*Biology, identification and control of plant root-knot nematode, Meliodogyne* ” North California State University Graphics Pp 22.
- Tibugari, H., Mombeshora, D., Mandumbu, R., Karavina, C. and Parwada, C. A. (2012). Comparison of the Effectiveness of the Aqueous Extracts of Garlic, Castor Beans and Marigold in the Biocontrol of Root-Knot Nematodes in Tomato. *Journal of Agricultural Technology*, 8(2): 479-492.
- Tolno, E., Kobayashi, H., Ichizen, M., Esham, M. and Balde, B.S. (2016). Potato production and supply by smallholder farmers in Guinea: An economic analysis. *Asian Journal of Agricultural Extension and Economics Society*, 8(3):1–16.