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Response of Root Knot Nematode to Three Systemic Herbicides and Their Effect on the Growth and Yield of Cowpea Grown in Umuahia South LGA, Abia State

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Abstract
The effect of herbicides (pendimethaline, imazamox and atarazine) on root-knot nematode (*Meliondogynespp*) and yield of cowpea (*Vigna unguiculata*) were compared in the field and pot experiments (August-October 2021 and September-Novermber 2022). The pot experiment was arranged in a Completely randomized design using plastic buckets with four treatments replicated three times including the control. Each bucket was inoculated with 2,000 nematode eggs. The treatments included: two pre-emergence (Pendimethalin and atrazine) herbicides and one post-emergence herbicide (Imazamox). The pendimethaline, imazamox and atrazine were applied using a knapsack at 250ml/16l. Data collected included: grain yield (number of seeds per pod and weight of seeds), root parameter (fresh root weight and number of root nodules), nematode galls and population (gall index, number of eggs and number of juveniles). The field experiments (the two seasons) were conducted on land naturally infested with root-knot nematode and were laid out in a Randomized Complete Block Design with four treatments including the control (no application). Similar treatments in the pot experiment were used. The herbicides were applied at the same rate as in pots. Data similar to those in the pot experiment were taken. The data collected in the experiments were subjected to analysis of variance (ANOVA) and means were compared using the least significant difference (LSD) at a 5% probability level ($P<0.05$). Results obtained at the end of the experiments showed that pendimethalin significantly reduced the nematode population and increased the yield of cowpea.
Keywords:* *Root knot nematode, systemic herbicides, cowpea

Introduction

Cowpea (*Vigna unguiculata*), is an annual herbaceous legume cultivated for its edible seeds for fodder cultivated cowpeas are herbaceous annuals that are either erect, prostrate or climbing annuals with a tap root and virtually all are glabrous (Horn, Nghituwamahata, and Ueitiele. 2022). It is a self-pollinated crop and can grow up to 80 cm and up to 2 m for climbing cultivars. Cowpea is a multipurpose grain legume; in which the entire plant can be used for either human or livestock consumption (Sarr, Bodian, and Leclerc, 2022). It is one of the most important food and forage legumes in the semi-arid tropics that include parts of Asia, Africa, Southern Europe, Southern United States, and Central and South America (Alemu *et al*., 2016).

Several plant-parasitic nematodes are associated with legume crops limiting growth and yield of cowpea (Gregory *et al*, 2017). Damage caused by PPNs has been estimated from $US80 billion to $US157 billion per year. However, the full extent of nematode damage is likely underestimated as many growers, particularly in developing countries, are unaware of the presence of PPNs (Jones *et al.*, 2013).

These nematodes are usually small soil-borne pathogens that can feed on all plant parts (including roots, stems, leaves, flowers and seeds), although most species feed on roots. The stylet is connected to three to five pharyngeal glands that produce effector molecules, which are often secreted, facilitating penetration, internal migration, and parasitism (Mejias *et al*., 2019). Based on their feeding habits, plant-parasitic nematodes can be broadly divided into ectoparasitic and endoparasitic. However, the most important nematodes in terms of crop losses are sedimentary endoparasites, the root-knot (*Meloidogyne spp*) and cyst nematodes (*Heterodera* and *Globodera* spp.) (Jones *et al*., 2013).Given the high economic impact caused by parasitic nematodes, a large number of strategies have been developed for nematode control in agriculture.

The use of herbicides has continued to be the dominant way of controlling weeds affecting crop production (Ofusu, Evan, Adrien, Gyory, Janox, and Gabriella, 2023). The effect of herbicide application on soil pathogens such as plant-parasitic nematodes has not been fully investigated. Strategies used to manage root-knot nematodes in tropical regions include nematicides, crop rotation, soil solarization, flooding, biological control, and soil amendments such as the application of manure, plant material, compost, and wood ash (Poudel, Budhathoki, Thapa, Mehta, Bhandari, and Shrestha, (2023).

Estango *et al.* (2016) hinted that some herbicides, such as pendimethalin and metholachlor, had significant negative effects on soil nematode populations, resulting in reduced abundance and diversity and other herbicides, such as imazethapyr and nicosulfuron, exhibited no significant direct effects on nematode populations. There is a need for further investigation to see if the herbicides applied by the farmers to control weeds could have an effect on plant parasitic nematodes in the soil. The objectives of the study were**:** to determine the influence of pre-emergence and post-emergence herbicides on cowpea yield and root-knot nematode population.

Materials and Methods

The experiment was conducted at Amakama Ikputu, Umuahia South LGA, Abia State, Nigeria. A small seeded variety of Cowpea (*Vigna unguiculata*) was obtained from NRCRI Umudike, Abia state. The treatments included neem leaf, charcoal, ash, two pre-emergence (Pendimethalin and atrazine) herbicides and one post-emergence herbicide (Imazamox). Root-knot nematode eggs were obtained from a culture of nematode-infected root of Basella alba (*cylon spinach*). Galled root pieces of Basella alba (*cylon spinach*)containing egg masses were cut into pieces and placed in a beaker of 500ml capacity with 200 ml of 0.5% chlorox (sodium hypochlorite, NaOCL solution shaken vigorously by hand for 4 mins. This was done to dissolve the gelatinous matrix encasing the eggs. The solution was be poured through two nested sieves, 200 mesh (75um) and 500 mesh (25um). Eggs in the 500 mesh sieve were washed free of NaOCL solution with slow stream of cold water into a beaker containing 1L of water. The cut root in the original beaker was washed twice with water to obtain additional eggs. The number of eggs per 1ml of water was estimated by counting 3 samples of 1ml each using Domncaster's counting dish under an electronic stereomicroscope and a working mean of eggs/ml was estimated.

Pot experiment: The effect of herbicides on root-knot nematode (Meloidogyne spp) on cowpea

The experiment was laid out in Randomized Complete Block Design (RCBD) using plastic buckets with seven treatments replicated three times including control. Topsoil was collected from Micheal Okpra University of Agriculture Umudike farm. The soil mixture was moistened and put into a cut drum, covered and heated until it reached a temperature of about 80°C and maintained at this temperature for 20mins (Ononuju *et al*, 2014). After cooling down, 7kg of the soil was separately filled in each of the 21 plastic buckets. Cowpea seeds were sown three seed per bucket. Two weeks after, the plant was thinned down to a healthy seedling per pot and fertilizer (NPK 15-15-15) was applied at 156kg/ha (Polthanee *et al*; 2015). Three weeks after emergence, they were inoculated by pouring a volume of 20ml of the suspension containing 2,000 nematode eggs extracted by making a groove around it. Using a knapsack the pre-emergence herbicides were applied before the plant emerged while the post-emergence herbicide was applied after emergence at 250ml per 16l. The control consists of pots with nematode but no treatment application. The plants were watered daily and weeded as when due. The data collected after eight weeks were; the weight of pods, weight of seeds, fresh root weights, dry shoot weights (using a digital laboratory weighing balance), number of galls and counting number of nodules. Nematode population in the soil: 200ml of soil samples were taken from each plant stand. Nematodes were extracted from soil samples using the modified Baermsn technique (Hopper, 1969). Nematode counts were made after 24hrs using a stereoscopic microscope (Ononuju and Nzenwa 2011). Nematode population in root: nematode eggs in the roots were estimated by measuring out 4.0g each of root samples of the plant infested with nematode eggs. Galled root samples were cut into small pieces and placed in a beaker of 500 ml, and thereafter processed as previously described in the extraction of nematode eggs from soil (Ononuju and Nzenwa 2011). Root-knot index: Gall rating was obtained by observing the number of galls on the roots of the cowpea plants after harvest. This was done on a 0-4 scale.

0-No infection (No gall present)

1-Rare infection (1-3 galls present)

2-Light infection (4-10 galls present)

3-Moderate infection (11-30 galls present)

4-Severe infection (more than 30 galls present) (Agu and Ogbuji, 1996)

Field experiment: The effect of herbicides on root-knot nematode (Meloidogyne spp) on cowpea

The field trial was conducted in a field that was naturally infested with root-knot nematodes and arranged in a randomized complete block design made up of six treatments and control replicated three times during (August-October 2021 and September-November 2022). The area was cleared after which the beds were made. Thereafter, the initial nematode population was determined using the modified Baermann technique (Trudgill and Phillips, 2020). Cowpea seeds were sown three seeds per hole in the field at 20 by 20 cm in each bed. Two weeks after, the plants were thinned down to a healthy seedling per stand.

The experiment was laid out in a randomized complete block design (RCBD) parameters measured were the same as described in the pot experiment. The data collected were subjected to Analysis of Variance (ANOVA) and means were compared using Least Significant Difference (LSD) at a 5% probability level**.**

Results and Discussion

Results

Effects of the treatments on pod and grain yield of cowpea infected with nematode in the pot experiment

Table 1 shows the effects of the treatments on the pod and grain yield of cowpea infected with nematode in the pot experiment. The pre-emergence herbicide 1 compared favourably with the post-emergence herbicide giving a better yield.

Effects of the treatments on dry shoot weight and nodulation of cowpea infected with nematode in the pot experiment

Table 2 presents the effects of the treatments on the dry shoot weight and number of nodules of cowpea infected with nematode in the pot experiment. No significant difference was observed among the treatments.

Effects of the treatments on galls and population of nematode in infected cowpea in the pot experiment

Table 3 presents the effects of the treatments on the number of galls, nematode eggs and the final number of juveniles in the soil in the pot experiment. All the treatments were compared favourably ($P \leq 0.05$) with the control and had fewer number of galls and nematode eggs. Furthermore, all the treatments except pre-emergence herbicide 2 treated plants compared favourably ($P \leq 0.05$)with the control, and had fewer juveniles in the soil.

Effects of the treatments on pod and grain yield of cowpea infected with nematode in the field during the 2021 and 2022 planting seasons

The effects of the treatments on pod and grain yield of cowpea infected with nematode in the field during the 2021 and 2022 planting seasons are shown in Table 4. In the 2021 planting season, the plants did not compare favourably with the control. But in the 2022 planting season, only the pre-emergence herbicide 2 treated plants had the greatest number of pods and compared favourably ($P \leq 0.05$) with the control. The pre-emergence herbicide 2 treated plants compared favourably ($P \leq 0.05$) with the control. They respectively had a greater number of seeds and an increased weight of seeds.

Effects of the treatments on dry shoot, root weight and nodulation of cowpea infected with nematode in the field during the 2021 and 2022 planting seasons

Table 5 is on the effects of the treatments on dry shoot, root weight and number of nodules of cowpea infected with nematode in the field during the 2021 and 2022 planting seasons. In 2022, only the post-emergence herbicide-treated plants compared favourably ($P \leq 0.05$) with the control. It however did not differ significantly from other treatments. On fresh root weight, no significant difference was observed between the treatments and the control 2021. However, plants treated with post-emergence herbicides weighed more and compared favourably ($P \leq 0.05$) with the plants treated with pre-emergence herbicides 2. In 2021, pre-emergence herbicide 2 treated plants had more nodules (3 respectively) while the pre-emergence herbicide 1 treated plants had the least number of nodules (1). Similarly in 2022, the post-emergence herbicide-treated plants had more number of nodules (5).

Effects of the treatments on galls and population of nematode in infected cowpea in the field during 2021 and 2022 planting seasons

The effects of the treatments on the number of galls and population of nematode in infected cowpea in the field during the 2021 and 2022 planting seasons are presented in Table 6. No significant difference was observed between the treatments and the control on the number of galls in the 2021 planting season. However, pre-emergence herbicides 1 and 2 treated plants had no galls respectively (0) while the control had more galls (4). On the other hand, in the 2022 planting season, all the treatments except post-emergence herbicide treated plants had fewer number of galls and differed significantly ($P \leq 0.05$) from the control. On the number of nematode eggs in the root, no significant difference was observed between the treatments and the control in 2021. But in 2022, all the treatments compared favourably ($P \leq 0.05$) with the control and had fewer number of nematode eggs. On the initial number of juveniles in the soil, in 2022, no significant difference was observed between the treatments and the control. On the final number of juveniles in the soil, no significant difference was observed between the treatments and the control in 2021. The post-emergence herbicide treated plants had the least final number of juveniles in the soil (33). However, in 2022, the pre-emergence 1 treated plants had the least number of juveniles and differed significantly ($P \leq 0.05$) from the control.

Discussion

In the pot experiment, plants treated with pre-emergence herbicides (pendimethalin and atrazine) demonstrated a significant difference compared to those treated with the post-emergence herbicide (imazamox). This distinction may arise from the indirect influence of herbicides on nematode populations, potentially through alterations in weed communities or the abundance of alternative hosts. Filho et al. (2019) supported this notion, finding that the herbicide glyphosate led to a significant reduction in weed biomass. Consequently, this decrease in weed population had a cascading effect, lowering the abundance of plant-parasitic nematodes that rely on weeds as hosts. Pre-emergence herbicide 2 had the highest number of final juveniles excluding the control. This could be a result of the inability of the herbicide to eradicate or reduce the weeds, concurrently reducing crop yield. This is supported by Ofusu et al. (2023) suggesting that weeds may encourage the development of diseases; provide shelter and act as an alternate host for pests (Poudel et al, 2023). In the first planting season, plants treated with pre-emergence herbicides displayed an absence of galls, while the control group exhibited 4 galls. Conversely, during the second season, plants treated with post-emergence herbicide had a significantly lower number of galls compared to the control. This variation could be attributed to the selective nature of certain herbicides towards specific nematode species or groups. Karimipour, Saeidi and Doryanizadeh, (2019) highlighted that certain herbicides can diminish the population of root-knot nematodes while fostering the growth of beneficial predatory nematodes. These selective effects may stem from differences in herbicide toxicity or the varying sensitivity of nematode species.

Conclusion

In all, the experiments showed that the application of herbicides to agricultural soil is important in the control of root-knot nematode disease on cowpea as well as enhance the growth and yield of the cowpea. However, the application of pre-emergence herbicides 1 and 2 significantly reduced the population of the root-knot nematode as well as improved the yield of cowpea respectively. Farmers are therefore encouraged to use pendimethalin in their cowpea farming because apart from controlling weeds it can also help in reduction of nematode population. Further research on rates of application of the herbicides both in green house and field trials are highly recommended so as to determine their best application rate.

Herbicide manufacturers are advised to do more research for proper dosages to incorporate reduction of nematode population as one of its benefits.

References

Alemu, M., Asfaw, Z., Woldu, Z., Amsalu, F., and Beth, M. (2016). Cowpea (*Vigna unguiculata* (L.)Walp.)(Fabaceae) landrace diversity in Northern Ethiopia. *International Journal of Biodiversity and Conservation*. 8(11): 297–309.

Estango, C. G., Pardales, J. R. Jr, and Ramos, M. R. Jr. (2016). Nematode response to metolachlor, pendimethalin, imazethapyr, and nicosulfuron in soybean–cowpea cropping system. *Philippine Agricultural Scientist*. 99(3): 237-246.

Filho, J. A., Souza, R. M., Silva, E. A., Alves, A. C., and Santos, D. F. (2019). Glyphosate and rust red rice control affect the community structure of nematodes in soybean. *RevistaCiênciaAgronômica*. 50(4): e199682.

Gregory C. Bernard, Marceline Egnin and Conrad Bonsi. (2017). The Impact of Plant-Parasitic Nematodes on Agriculture and Methods of Control. *Nematology - Concepts, Diagnosis and Control*. 7(61*)*

Horn, L., Nghituwamahata, S. N. and Ueitiele I. (2022).Cowpea Production Challenges and Contribution to Livelihood in Sub-Sahara Region. *Agricultural Sciences*. 13(01): Doi:10.4236/as.2022131003. license. CCBY4.0 Pp 25-32.

Jones, J. T., Haegeman, A., Danchin, E. G. J., Gaur, H. S., Helder, J., Jones, M. G. K., et al. (2013). Top 10 plant-parasitic nematodes in molecular plant pathology. *Molecular Plant Pathology*. 14, 946–961.

Jones, J. T., Haegeman, A., Danchin, E. G. J., Gaur, H. S., Helder, J., Jones, M. G. K., et al. (2013). Top 10 plant-parasitic nematodes in molecular plant pathology. *Molecular Plant Pathology*. 14, 946–961.

Karimipour Fard, H., Saeidi, K., & Doryanizadeh, N. (2019). Effects of chicken manure and summer ploughing on root-knot nematode (*Meloidogyne javanica*) management in cantaloupe (*Cucumis melo* var. *Cantalupensis*). *Archives of Phytopathology and Plant Protection*, 52(15–16), https://doi.org/10.1080/03235408.2019.1694384

Mejias J., Truong N. M., Abad P., Favery B., Quentin M. (2019). Plant proteins and processes targeted by parasitic nematode effectors. *Frontal Plant Science*. 10:9-70.

Ofusu, R., Evan, D. A., Adrien, M., Gyory, P., Janox, T. and Gabriella K. (2023). Herbicide Resistance: Managing weeds in a Changing World. *Agronomy.* 13(6), 1595; https://doi.org/10.3390/agronomy13061595. 23-36.

Ononuju, C. C. and Nzenwa, P. O. (2011). Nematicidal effect of some plant extracts on egg hatchability and control of *Meloidogyne spp*. in cowpea (*Vigna unguiculata (L.) Walp*). *African Journal of Plant Science*. 5 (3): 176-182

Polthanee, A., Promkhambut, A., Manaonok, J and taboonmuang, R. (2015). Pre-Rice or Post-Rice Mungbean Productivity with Chemical and Bio-Compost Fertilizer under Rainfed Conditions. *International Journal of Environmental and Rural Development*. 6(1): 138-142.

Poudel, S. R., Budhathoki, R., Thapa, A., Mehta, A., Bhandari, S., & Shrestha, S. M. (2023). Eco-Friendly Management of Root Knot Nematode (*Meloidogyne* spp.) in Okra (*Abelmoschus Esculentus*) Using Different Soil Amendments. *AgroEnvironmental Sustainability*. 1(3). https://doi.org/10.59983/s2023010302. Pp 210-218.

Sarr, A., Bodian, A. and Leclerc, C. (2022). Ethnobotanical study of cowpea (vigna unguiculata (L) Walp). *Journal of Ethnobiology and Ethnomedicine*. 6, 16-25.

Trudgill, D. L. and Phillips, M. S. (2020). Nematode Population Dynamics, Threshold Levels and estimation of crop losses. Fao.org/3/v9978/v99. P2.

Plate 1: Pot experiment showing inoculated cowpea plants treated with plant extracts and herbicides

Plate 2: The Field Trial during the First Planting Season, 2 weeks after planting. (August –October 2021)

Plate 3: The Field Trial during the Second Planting Season, 4 weeks after planting. (September –November 2022)

Table 1: Effects of the treatments on pod and grain yield of cowpea infected with nematode in the pot experiment

Treatment	**No of pods/stand**	**Weight of pods (g)**	**No of Seeds**	**Weight of seeds (g)**
Imazamox	3.00	9.60	13.00	9.10
Atrazine	3.33	11.50	13.70	10.90
Pendimethalin	2.00	5.30	9.30	6.50
Control	0.33	1.30	2.30	1.00
Mean	2.17	6.93	9.58	6.88
$LSD_{(0.05)}$	4.76	15.39	18.86	13.89

Field work 2021

Table 2: Effects of the treatments on dry shoot weight and nodulation of cowpea infected with nematode in the pot experiment

Treatment	**Dry Shoot Weight (g)**	**No of Nodules/stand**
Imazamox	9.70	2.00
Atrazine	12.10	1.33
Pendimethalin	12.60	5.00
Control	8.50	0.33
Mean	16.21	3.24
$LSD_{(0.05)}$	10.59	NS

Field work 2021

Table 3: Effects of the treatments on galls and population of nematode in infected cowpea in the pot experiment

Treatments	Number of galls	No of Nematode Eggs	Final No of Juveniles in the soil
Imazamox	1.67	167.00	3.00
Atrazine	0.00	0.00	33.00
Pendimethalin	1.00	67.00	600.00
Control	7.00	567.00	767.00
Mean	1.38	114.43	224.43
$LSD_{(0.05)}$	2.83	175.10	238.70

Table 4: Effects of the treatments on pod and grain yield of cowpea infected with nematode in the field during 2021 and 2022 planting seasons

	No of pods/ Net plot		Weight of pods (g)		No of Seeds /Net plot		Weight of seeds (g)	
Treatment	**2021**	**2022**	**2021**	**2022**	**2021**	**2022**	**2021**	**2022**
Imazamox	14.00	18.00	41.70	50.30	96.00	69.30	22.60	45.80
Atrazine	10.70	20.70	26.40	38.90	79.00	82.70	18.60	36.10
Pendimethalin	17.70	47.30	49.10	105.60	177.00	183.30	29.40	96.50
Control	13.70	17.30	31.80	22.10	38.00	53.30	36.00	18.10
Mean	21.03	5.23	59.00	63.06	175.00	103.13	41.27	56.79
$LSD_{(0.05)}$	18.31	20.46	39.56	56.13	182.90	76.38	29.62	53.46

Field work 2021/2022

Table 5: Effects of the treatments on dry shoot, root weight and nodulation of cowpea infected with nematode in the field during 2021 and 2022 planting seasons

	Dry Shoot Weight (g)		Fresh Root Weight (g)		No of Nodules	
Treatment	**2021**	**2022**	**2021**	**2022**	**2021**	**2022**
Imazamox	73.40	50.90	12.27	15.10	1.67	5.33
Atrazine	78.60	22.00	9.27	18.10	1.00	1.67
Pendimethalin	82.10	23.90	7.43	19.33	3.00	2.00
Control	56.50	15.60	9.63	16.13	1.33	1.33
Mean	77.26	30.59	9.31	14.91	2.09	2.48
$LSD_{(0.05)}$	34.60	29.49	3.81	6.29	NS	NS

Table 6: Effects of the treatments on galls and population of nematode in infected cowpea in the field during 2021 and 2022 planting seasons

	Number of galls		No of Nematode Eggs		Initial No of Juveniles in the soil		Final No of Juveniles in the soil	
Treatment	**2021**	**2022**	**2021**	**2022**	**2021**	**2022**	**2021**	**2022**
Imazamox	3.33	5.67	367.00	500.00	1100.00	2000.00	33.00	400.00
Atrazine	0.00	3.33	133.00	367.00	900.00	1200.00	167.00	167.00
Pendimethalin	0.00	0.33	200.00	33.00	1000.00	1433.00	100.00	433.00
Control	4.33	10.00	733.00	1500.00	733.00	1833.00	100.00	867.00
Mean	1.95	3.24	299.86	247.57	1028.43	514.14	128.57	371.43
$LSD_{(0.05)}$	NS	6.09	NS	741.30	792.00	NS	NS	584.50

Field work 2021/2022