Journal of Agriculture and Environment Vol. 19 No. 1, 2023: 183-188
ISSN: 1595-465X (Print) 2695-236X (Online)

IN VITRO EFFICACY OF SWEET ORANGE PEELINGS EXTRACT ON EGG HATCHABILITY AND JUVENILE MORTALITY OF *Meloidogyne javanica*

M.Y. Adamu

Department of Crop Protection, Modibbo Adama University Yola

ABSTRACT

The effect of extract of Sweet Orange Peeling (SOP) was investigated on egg hatchability and juvenile mortality of *Meloidogyne javanica* in the laboratory. One hundred (100) eggs were introduced into each of 15 Petri dishes and 1000 juveniles in 15 Petri dishes each were independently exposed to crude extract (100%) concentration, crude + 5m1 distilled water crude + l0ml distilled water, crude + 15m1 distilled and distilled water only served as control. The Petri dishes were arranged in completely randomized design, percentage egg hatchability and mortality were calculated over l20 hours. The results indicated that crude extracts gave the least percentage of egg hatching with highest percentage of juvenile mortality. Other dilutions also significantly inhibited egg hatching and caused juvenile mortality when compared to the control. The results further revealed that hatching decreased with increase in duration of exposure and juvenile mortality increased with increase in exposure time. It could therefore be concluded that extracts were able to reduce egg hatching and cause juvenile mortality of *M. javanica* in the laboratory. However, further research in screen house and field is recommended to ascertain their efficacy.

Keywords: Sweet orange peelings; extracts; juvenile; hatchability; mortality

INTRODUCTION

Plant materials are important sources of naturally occurring pesticides and/or nematicides as many phytochemical compounds with nematicidal potential or activity have been found in them (Chitwood, 2002). These phytochemicals include alkaloids, tannin, saponin, flavonoid, diterpens, fatty acids, glycocides, sesquiterpenes, thienyls, Isothiocyanate and limonene among others. These plant materials will kill or repel pest, disrupt their life cycle and even discourage them from feeding, and this, in turn, affect the development and reproduction of many plant parasitic nematodes. Various plants extract like *Eucalyptus* species *Avicennia manna, Rhizophora mucronate, Ceriops tagal* and *Acgicera's corniculatum*, mangold, rice husk, neem cake and powder extract, garlic power extracts, sweet orange peeling, white brassica and molasses have been reported to have pesticidal properties. Tsay *et al.* (2004) reported that tissue extract of Indian blanket was lethal to mobile juvenile of *M. incognita* and were inhibitory to the hatching of eggs at 250ppm. Umar (2003) also reported that 100% concentration of extract of *Nicotiana tabaccum* and *Syzgnum aromalicum* causes 100% mortality of *M javanica* in the laboratory.

Root-knot nematode, *M. javanica* (kofoid and white) Chitwood is one of the major plant parasitic nematodes species that have been regarded as the greatest problem of food production in Nigeria and other developing countries, where they have great impact on crop productivity. *M* species are difficult to control because of their diverse host range and high rates of production. They induce major morphological and physiological changes within the root when attacked. The use of synthetic nematicides, considered the most effective practical means of combating plant parasitic nematodes, as they have been found to increase crop yield. However, they are expensive, and also lead to environmental hazard because of high toxicity and persistence, toxic to human and other beneficial organisms. Therefore, the need to develop alternative non synthetic nematicides that are ecofriendly and effective in controlling plant parasitic nematodes (Nchore *et al.*, 2011). In view of this, the study is aimed at investigating the effect of different concentrations of sweet orange peelings extracts on egg hatchability and juvenile mortability of *M. javanica* in the Laboratory.

MATERIALS AND METHODS

Experimental Site

The experiment was conducted in the Plant Pathology Laboratory of the Department of Crop Protection, Modibbo Adama University, Yola, Adamawa State, Nigeria. Yola is located within latitude 9° 9'N and longitude 12°30'E at an altitude of 185.9m above sea level (Bashir, 2000).

Preparation of Sweet Orange Extract

Sweet orange peelings were collected from Jimeta ultra-modern market of Adamawa State, washed and Shed-dried for seven days on polythene sheets. The dried material was then grounded into powder using pestle and mortar (Kasvala, 2007). From the grounded powder, 40g was soaked in conical flask containing 100ml of distilled water and left for 24-48hrs and then filtered through What Mann, No.2 filter paper. The filtrates so obtained constitute 100% undiluted concentration otherwise designated as crude extracts (C). This was then diluted with 5, 10 and 15m1 of distilled water, to give concentrations C1, C2, C3 respectively. Distilled water was used as control.

Phytochemical Analysis of Sweet Orange Extracts

Chemical tests for tannins, saponin, flavonoids, Glycocides and alkaloids were carried out in the Laboratory on the plant extracts using the method described by Sofowora (1993).

Extraction of *M. javanica* Juveniles (J2)

M. javanica juveniles were extracted from pure infested galled tomato roots using the Modified Baermann funnel method (Whitehead and Hemming, 1965). The juveniles were viewed and identified under an Electron Microscope.

Extraction of Nematodes Eggs

Egg masses of *M. javanica*, from the roots of infested tomato plants were used to prepare nematode egg suspension. Nematode eggs were extracted by vigorously shaking of the tomato root in 0.005% sodium hypochioride for two (2) minutes (Hussey and Barker, 1973). Eggs were collected and rinsed with tap water through a 75 cm sieve and collected on a 26 cm sieve, then transferred into distilled water forming the egg suspension (Dong *et al.*, 2007).

Egg Hatching Test

Ten (10) ml of the crude extract and its 5, 10, l5ml diluted forms were separately dispensed into the Petri dishes containing appropriately 100 eggs/ml. Petri dishes for the control contained distilled water only (no extracts). Five treatments replicated three times; Petri dishes were arranged in Completely Randomized Design in the Laboratory. Eggs hatched were observed microscopically and the mean percentage of egg hatching was collected over a period of 120 hours.

Juvenile Mortality Test

Aliquots of l0ml each of crude extracts and its 5, 10, 15ml diluted forms of sweet orange peelings, were separately dispensed into the Petri dishes containing approximately 1000 second stage juvenile (J2) of *M. javanica* while Petri dishes for control contained distilled water only (no extract). Five treatments replicated three times arranged in Completely Randomized Design (CRD). Dead nematodes were identified by touching them with a needle under microscope whether they exhibit mobility or not. Nematodes were considered dead when immobile, shrinked or internally vacuolated. Mean juvenile mortality was observed over a period of 120 hrs.

Data Analysis

All data collected were subjected to Analysis of Variance and means separation was done using the Duncan's New Multiple Range Test at 5% level of probability.

RESULTS

The results of the phytochemical analysis of sweet orange peelings extract indicated the moderate presence of saponin and alkaloids, high presence of tannins, flavonoids and Glycocides (Table 1). The result on egg hatching revealed that crude extract (100%) concentration gave the highest inhibition of egg hatching (10%) at l20 hours, closely followed by 5 m1 diluted form (11%). A significant reduction of eggs hatching was also observed when the crude extracts were diluted with load 15m1 of distilled water. The least percentage inhibition was observed in the control (85%) with no extracts. The results further revealed that, as the extracts dilution increases, the percentage eggs hatching decreases, also eggs hatching decreases with an increase in exposure time. The result on juvenile mortality also revealed that crude extracts 100% concentration gave the highest juvenile mortality (90%), other concentrations were also effective as significant juvenile mortality was recorded as

89.2, 87.5 and 84.9% for 5, 10, and 15 ml respectively. The control recorded zero juvenile mortality (Table 2) at l20hours. The results further revealed that juvenile mortality increases with increased in duration of exposure, as also extract diluted mortality found to be decreasing.

Table 1: Phytochemical analysis of the plant materials

Phytochemical	Presence
Glycocides	+++
Flavonoids	+++
Tannins	+++
Alkaloids	++
Saponnis	++

++ = Moderately present
+++ = Highly present

Table 2: Effect of sweet orange peelings extracts on egg hatching and juveniles mortality of *M. javanica* at 120 Hours exposure time

Treatments	Hatching % at 120 hours	Number of Juvenile eggs	Mortality % at l20 hours
C_1	10	100	90.1
C_2	11	100	89.2
C_3	12	100	*87.5*
C_4	14	100	84.9
C_5	85	100	0.00

C_1 Crude Extract, C_2 Crude Extract + 5ml Distilled water,
C_3 = Crude Extract + 10ml Distilled water, C_4 Crude Extract + 15ml Distilled water,
C_5 = Control (Distilled Water only).

DISCUSSION

The best nematicidal activity shown by 100% concentration (crude extract) as a result of the maximum inhibition of eggs hatching and highest juvenile mortality of *M. javanica* were recorded. The nematicidal effect of the tested plant extract could be attributed to the presence of high content of phytochemical compounds which has the ability to dissolve the cytoplasmic membrane of nematodes cells, thereby interfering with enzyme protein structure (Knoblock *et al.*, 1989). This type of control effects of citrus peels extracts on plant parasitic nematode have been reported by many researchers including Abolusoro (2010); Izuogu *et al.* (2019); Mohanapriya and Ragini (2023). Other treatments though significant but were less effective as compared to the crude extract (100% concentration). This result corroborated that of Umar (2013) who reported that 100% concentration of extracts of *Nicotiana labacum* and *Syzgnum aromalicum* caused 100% juvenile mortality of *M. javanica* in the Laboratory. The maximum inhibition of egg hatching, and highest juvenile mortality of the extract could be due to the presence of phytochemical with nematicidal activity such as tannins, saponnins, flavonoids, glycocides, alkaloids, that affected the Embryonic development, killed, the juveniles or the egg even dissolve the egg masses. This corroborates with the findings of Eman and El-nuby (2019) whose study revealed that plant extracts containing flavonoids,

lipids, saponins, amides, glycocides, limonene singly or in combination inhibited egg hatching.

The control Petri dishes been water only recorded the highest number eggs hatched and zero mortality. This was due to the fact that the control Petri dishes contained no extracts hence no phytochemicals in them resulted in high number of hatched eggs with no death of nematodes (Jacob *et al*., 2021).

CONCLUSION

The study indicated that sweet orange peelings extract has nematicidal property as applied crude concentration and its diluted forms were also effective in inhibiting eggs hatching and causing juvenile mortality of *M. javanica* in the Laboratory. It is suggested that field trials are conducted to determine the level of effectiveness of the extract before recommendations are made to the farmers in the study area.

REFERENCES

Abolusoro, S.A. Oyedunmade, E.A. and Olabiyi T.I. (2010). Evaluation of sweet orange peel aqueous extract (*Citrus sinensis*) as root –knot nematode suppressant. *Journal of Tropical Agriculture, Food, Environment and Extension*, 9(3): 170 – 175.

Adegbite, A.A. and Adesiyan, S.O. (2005). Root extracts of plants to control root-knot nematode on edible soybean. *World Journal of Agricultural Science*, (1): 18-21.

Adegbite, A.A. (2003). Comparative effects of Carbofuran and water extract of *Chromolaena odorata* on growth, yield and food components of root-knot nematode-infested soybean *(Glycine max* L.) Merril. Ph.D. Dissertation University of Ibadan, Ibadan, Nigeria.

Agyarko, K. and Asante, J.S. (2005). Nematode dynamics in Soil amended with Neem leaves and Poultry manure. *Asian Journal of Plant Science*, 4(4): 426 - 428.

Bashirs A. B. (2000). Water quality and outbreak of diseases in Yola, Adamawa State, M.Sc. Thesis submitted to the Department of Geography, Federal University of Technology, Yola, l20p.

Chitwood, B. (2002). Phytochemical based strategies for nematode control. *Annual review of phytopathology*, 40(2): 21- 49

Dong, W., Holbrook, C.C., Timper, P., Brenneman, B.T. and Mullinix, B.G. (2007). Comparison of methods for assessing resistance to *Meloidogyne arenaria* in peanuts. *Journal of Nematology*, 39 (2): 169 - 175.

Eman, A.A. and El-Nuby A.S.M. (2019). Phytochemical and antinematodal screening on water extracts of some plant wastes against meloidogyne incognita. *International Journal of Chemical and Pharmaceutical Sciences*, 10(4)

Hussey, R.S. and Barker, K.R. (1973). A comparison of methods of collecting inocula of *Meloidogyne spp.* including a new technique. *Plant Disease Report*, 57: 1025-1025.

Izuogu, N.B., Azubuike, C. and Akeem, O. (2019). Comparative evaluation of powdered and aqueous extract of citrus peels and carbofuran in the management of root-knot nematode on infected cowpea field. *Agric. Conspec. Sci.*, 84(2): 173-178.

Kaskavala, O. (2007). Effect of soil solarization and organic amendments treatments for controlling *Meloidogyne incognita* in tomato cultivars in Western. *Anatolia Turkish Agriculture Forum*, 31: 159 167.

Jacob, M., Salu, C., Tochukwu, O. and A.A. Okwoli (2021). Effect of *Tamarindus indica* L. leaf extract and *Citrus sinensis* L. Osbeck peel extract on *Meloidogyne* spp (Root Knot Nematodes) in Jos, Plateau State. *International Journal of Pathogen Research*, 7(4): 15-21.

Knoblock, K., Weis, K. and Wergent, R. (1989). Mechanism of antimicrobial activity of essential oils. Proceedings of 37

Mohanapriya, B. and Ragini, G. (2023). Nematicidal activity of fruit citrus peels against root-knot nematode. *International Journal of Advanced Research in Biological Sciences*, 10(4): 215-230.

Nchore, S.B., Waceke, J.W. and Kariuki, G.M. (2011). Use of agro-industrial waste and organic amendments in managing root- knot nematodes in black night shade in selected parts of Kenya. *In*: 10th African Crop Science Conference proceedings, October 10 -13, 2011, Maputo, Mozambique. pp187-193.

Sofowara, A. (1993). *Medicinal Plants and Traditional Medicine in Africa*. Spectrum Books Ltd. Ibadan, Nigeria P. 289.

Tsay, T.T., Wu, S.T. and Lin Y.Y. (2004). Evaluation of *Asieraceae* plants for control of *Meloidogyne incognita*. *Journal of Nematology*, 36 (1): 36 -41.

Umar, I. (2013). Nematicidal potential of *Syzgbum aromaticum* extract on *M javanica* on tomato. *Taraba Journal of Agricultural Science*, 1(1): 97-99.