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ENHANCEMENT OF TOMATO ROOT-KNOT NEMATODES CONTROL USING A NOVEL BIOLOGICAL TREATMENT IN NORTH SINAI

Darin M.R. El- Bolok[1], Ahmed.A.ElSharawy[2]*, Eman K. Din[2] and M.Y. Abdalla[3]

[1] Environmental protection Department, Faculty of Environmental Agricultural Science, Arish University, Al Arish- 45516, Egypt.

[2] Plant Pathology Lab., Plant Production Department, Faculty of Environmental Agricultural Science, Arish University, Al Arish- 45516, Egypt.

[3] Faculty of Desert and Environmental Agriculture, Matrouh University, Egypt.

*Corresponding author: Dr. Ahmed. A. ElSharawy,

E-mail: Ahmed_Plantpathology@hotmail.com

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ABSTRACT

This study focuses on using olive mill waste water (OMWW) as a novel biological treatment to decrease plant diseases while improving food quality. Root-knot nematode *Meloidogyne* is considered one of the main tomato diseases in North Sinai. The current study aimed to evaluate the efficiency of diffracting olive mill waste waters concentrations compared with the nematicide Oxamylon on Tomato Root-knot. OMWW was applied daily to tomato plants growing in sandy loam soil for three months. Represented five different concentration of OMWW (0%, 25%, 50%, 75%, and 100%) used for irrigation during the experiment. The positive control treatment was treated with Oxamyl at recommended dose. The negative control was watered only with tap water (0%). The results showed that mortality rates of *M. incognita* were significantly affected by concentrations. All treatments were superior to the untreated control in reducing nematode, except for OMWWs at 100 **%**, which was equivalent to the control treatment. The largest drop in root galling numbers Oxamyl was achieved (75.73%) was followed by 50% at (73.81%), 75% (68.01%), and 25% (39.67%). The highest reductions rated were recorded with the 50%and 75% concentrations used of OMWW. Overall, in the effort of efficient biological treatments was achieved for root knot control measures in tomato, the OMWW application may be used as one factor of integrated root-knot nematode management.

***Keywords* :** Root-knot nematode, olive mill waste waters, Oxamyl.

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is one of the world's most popular crops, heavily consumed because of its high sensory appeal and desirable appearance (Casals *et al*., 2011). Egypt, is considered one of the five leading producers of tomato in the world (Pino *et al*., 2017). In 2017, The total area under tomatoes production in Egypt was approximately 7.3 million tons. In Egypt,Crop losses due to the nematodes infection on 80 crops, 15 of which are ‘life sustaining’, were estimated at L.E. 15.9 (= $2.30) billion annually based on 2011-2012 Egyptian production (Abd-Elgawad, 2014). Tomato is attacked by several biotic stresses, especially fungi and phytoparasitic nematodes. Plant-parasitic nematodes (PPNs) is one of the most globally inflicting damage in tomato plant(Abd-Elgawad, 2014; Abd-Elgawad, 2019).

The most important pathogens present in the roots of tomato plants are known to be root-knot nematodes (*Meloidogyne sp.*) (Moens *et al*., 2009). Additionally, it attack a wide variety of solanaceous crops causing serious economic losses *Meloidogyne species* can attack over 3000 plants (Mai, 1985; Mitkowski and Abawi, 2003). Root-knot nematodes comprises more than 98 species throughout the world (Abad *et al*., 2003; Jones *et al*., 2013; Taylor and Sasser, 1978). *Meloidogyne* species causes plant yield suppression, chlorosis, spreading wilting, root galling and leaf nutritional deficiencies (Abd-Elgawad, 2021).

Nowadays, researchers are searching for alternatives for methyl bromide (MeBr) which has been banned since 2005 as it has been identified as one of the chemicals that shared in the ozone depletion (Stromberger *et al*., 2005).

Olive mill waste water could be used in biological management of plant diseases, since it contains significant amounts of antimicrobial and phytotoxic compounds especially polyphenols (Alfano *et al*., 2011). During oil extraction from olive large volumes of unwanted olive mill wastewater (OMWW) produced, which is generated annually between October and December (Al-Khatib *et al*., 2009).

North Sinai is one of Egypt's governorate known for the high production of olive and olive oil, and during the production of olive oil tons of olive oil manufacturing waste are produced and need disposal (Abouseoud *et al*., 2008; Stanghellini and Miller, 1997).

One alternative that has been recommended to decrease the incidence of plant diseases is the use of olive mill wastewater (OMWW) (Yay *et al*., 2012). This method can be employed in both conventional and biological agricultural systems for control nematodes (Bonanomi *et al*., 2006; Paredes *et al*., 1999). According to several studies, olive residue proved to have phytotoxic and antimicrobial impacts (Martın *et al*., 2002; Moreno *et al*., 1987; Perez *et al*., 1992).

Roig *et al*. (Roig *et al*., 2006) developed a method for phenol degradation of liquid olive residues. Phenols are considered the main molecule in the OMWW reposable of antimicrobial and phytotoxic effects (Hadrami *et al*., 2004; Marrara *et al*., 2002; Mekki *et al*., 2006). These properties of OMWW might be used for suppression of plant pathogen, which will create recycling opportunities for these rare bioactive by-products (Capasso *et al*., 1995; Isidori *et al*., 2005).

The aim of this research was to study the effects of OMWW at different concentrations as a mechanism for the control of tomato root-knot nematode in North Sinai. Additionally, determine the effects of OMWW compared with a certain chemical nematicide (Oxamyl), which can be adopted for eliminating possible environmental problems associated with uncontrolled disposal of OMWW.

MATERIALS AND METHODS

Collection of Olive Mill Waste Water (OMWW)

The OMWW was obtained from a 3-phase olive mill plant at Al-Arish, North Sinai, Egypt. This experiment was performed in 2018-2019 at the Plant Pathology Lab., Department of Plant Production at Arish University. The OMWW was stored in tightly closed PVC vessels at room temperature until use.

Field studies

Tomato planting:

Plants of *Solanum lycopersicum* cv. “Super strain B” was used for the agronomical experiment to study the useof OMWW for the control of *Meloidogyne incognita* under controlled conditions. Fifteen seeds were planted in each pot and covered with a thin layer of soil. The pots were split out into five sections.All pots were put in 3 rows and randomly dispersed and labeled with 5 colors representing different concentrations to make irrigation with the various dilutions of OMWW simple. Irrigation was carried out using both the pointed OMWW dilutions and the control treatment using Tabwater. The plants were first irrigated for one week with tap water. Then 100 ml of solution with different OMWW concentrations (0%, 25%, 50%, 75% and 100 %) were usedfor each treatment, this process was replicated every day under greenhouse conditions, with temperatures varying from 25-35 ^{o}C during the day and 20-27 ^{o}C during the night. Pots that were irrigated with a series of dilutions of OMWW (0%,25%, 50%, 75%and 100%and control plants were watered with tapwater. This experiment was conducted in two consecutive years and each treatment consisted of threereplications.

A comparison treatment with the nematicide Oxamyl 24% was conducted. Application rate for the chemical nematicide Oxamyl was 1.5 gm/I water (Mennatoullah *et al*., 2010). This treatment hadthreereplications. The tomato plant length of shoots and root, fresh and dry weight (g) for both shoots and roots as well as fruit fresh weight were recorded. Plants were dried in an oven to a constant weight. These weights were recorded as g/plant for both shoots and roots (Abd El-Samad *et al*., 2019).

Statistical Analysis

A non-parametric test (Kruskal-Wallis variance analysis (ANOVA) on ranks) was used to compare the multiple therapies, as the data had unequal variances (statistical software Sigma stat 9). To isolate variations between treatments, the Student Newman Keuls Procedure was used.

RESULTS AND DISCUSSION

Effect of different concentrations of OMWW on Root-Knot nematodes

The goal of our studywas to determine the viability of using various concentrations of olive mill waste water OMWW (i.e. 0%, 25%, 50%,75% and 100%) on root-knot nematode of tomato. Oxomyl was used as a positive control besides an untreated 0 percent as a negative control.

In our experiment, tomato plants started showing signs of infection, including wilting and stunting as well as root galls.showed that the effect of OMWW against root-knot nematode was significantly different compared tocontrols. In both seasons, relative to untreated control. The findings obtained in both seasons were statistically. Figure 1. Presented that irrigation with 50% resulted in significant reduction in root-knot nematode with 73.8%. Furthermore, iirrigation with 100 % OMWW least effective treatment in suppressing the root-knot nematode with 15.5%. Another explanation is that certain polyphenols that have antiseptic activities and antioxidant properties could be found in the filtrate. Comparing the Oxamyl nematicide with the 0%treatment, it seems that Oxamyl greatly decreased the development of nematodes in both seasons by 75.73%.

In the first season, comparing the Oxamyl treatment with the five concentrations of OMWW using soil drenching significantly decreased the incidence of root-knot nematode on tomato. Among these five concentration 50% and 75% provided significant control of the disease, by 73.81 and 68.01%, respectively. The other two concentrations (25% and 100), including foliar application, were less effective, with the control ranging from 39.67to 15.47% respectively. Foliar applications of Oxamyl at 1.5 mg/mL was phytotoxic, causing the edges of leaves to curl up. Efficient control of root-knot nematode in agricultural systems by any chemical or biological compound depends on the mode of action by which the agent causes permanent or temporary damage to the cycle of nematode (Hausbeck and Lamour, 2004). Our experiments on suppression of root-knot nematode disease of tomato using OMWW showed that the foliar application of Oxamyl 1.5 mg has historically been used to control root-knot nematode on tomato plants (Landi *et al*., 2018; Radwan *et al*., 2012).

Fig. 2 : Effect of OMWW on different vegetative tomato growth parameters in terms of (A) shoot, root lengths, (B) shoot, root fresh weights, (C)shoot, root dry weights.

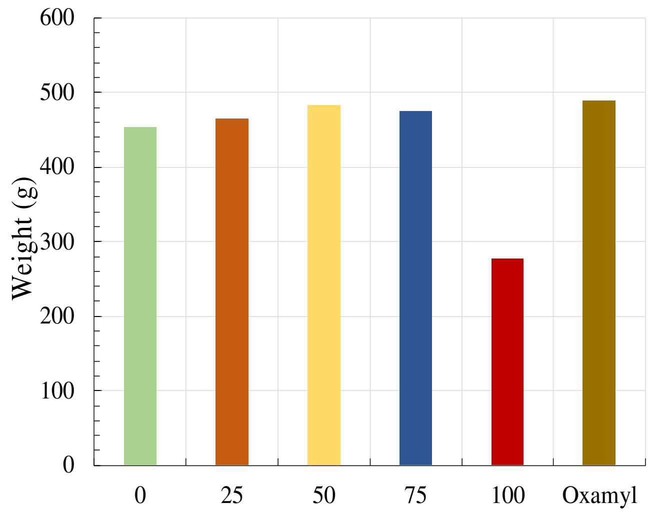

Fig. 3 : Effect of OMWW concentration (%) on of shoot, root fresh fruit weight.

The extensive suppression of using tomato root-knot nematode OMWW indicates the presence of bioactive compounds capable of inhibiting nematode population. This strong antagonistic action may play a key role in the suppression of pathogens, thus interrupting the life cycle of the nematode. Chitwood *et al.* (2002) reported among the nematicidal-capable phytochemical compounds were phenolics (pyrocatechol, caffeic acid and vanillic acid) and fatty acid derivatives (Chitwood, 2002). This fact seems to be confirmed in this study, where the least inhibition was provided by sample, which had the lowest OMWW concentration. However, with increasing dilution, the inhibition was decreased. The non-inhibitory reaction of 0 concentrations is in line with the recorded findings by Nico (Nico *et al.*, 2004), who found limited nematotoxic activity of OMWW, suggesting that the decomposition process in irrigation eliminates the inhibitors. Although ideal for plants, it is also not capable of managing root-knot nematodes in potting mixtures. Based on the initial dosage, they observed various responses and indicated that the inhibition was linked to nematotoxic compounds emitted from the concentration content (tannins or phenolic compounds) and was not attributed to microbial activity enhancement.

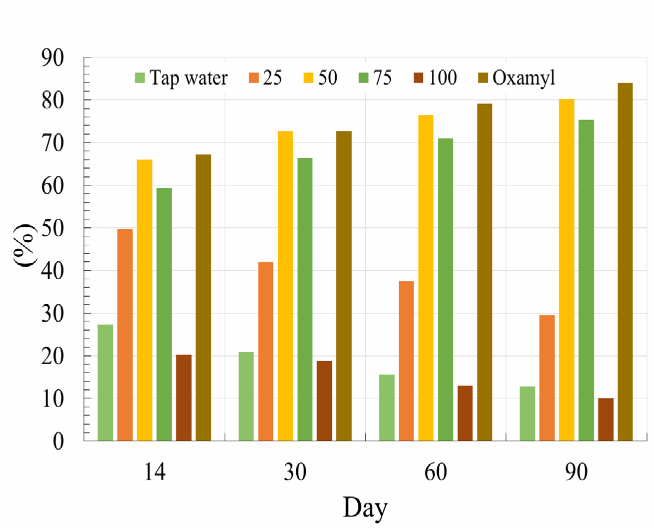

Fig. 1 : Effect of different concentrations of OMWW on Root-Knot nematodes in sandy loam soil for 14, 30, 60 days.

Effect of OMWW concentration (%) on vegetative tomato growth parameters in terms of shoot, root lengths, fresh, dry shoot weights and fresh fruit weight

Experimental results revealed in Fig. 2 that, Values were significantly different (P ≤ 0.05) among the different concentrations of OMWW. The plant height shoot and root length was decreased significantly with increasing the OMWW concentrations, The overall increasing values for both roots and shots lengths were recorded with the Oxamyl treatment and 50% OMWW (34.93; 31.13 cm and 59.26 : 57.45cm), respectively, however, the lowest was obtained with untreated control treatment was calculated Figure 2 (A). A Similar inclination was observed with another measured variable for both shoots and roots fresh and dry weights of tomato, when OMWW was used at 50% recorded 281.39 and 74.39 g/plant for both shoots and roots with percentage increase of 191.63 and 55.79 g/plant over untreated control treatment, respectively. Regarding both dry of both stem and root, the higher treatment recorded 95.37:35.56 and 93.86:34.15 g/plant it was obtained Oxamyl treatment and 50% OMWW Figure 2 (B,C).

In Figure 3. Comparing Oxamyl treatment with OMWW concentrations treatment in fresh fruit weight, it appears that the Oxamyl treatment significantly increasing fresh fruit weight by 488.59 g/plant during the fresh growing season. In addition, the untreated control was the least effective treatment in suppressing the fresh fruit weight with 278 g/plant reduction. The overall increases in fresh fruit weight were found with 50% and 75% OMWW treatment which recorded 482.37 and 474.80 g/plant respectively.

The present thesis research the use of OMWW for the management of root-knot nematode on tomato, with the second objective of evaluating the impact of various concentrations on the OMWW root-knot nematode population density.

These data are in agreement with Telles (Telles *et al.*, 2017), who studied the phenolic compound profiles and their interactions with the bean's protective mechanisms against the contamination of tomato root-knot nematode and the inhibitory action of amylase from nematode. They reported that phenolic compounds can potentially inhibit the attack of nematodes., Mendes *et al.* stated that numerous biotic and abiotic influences, such as the phenolic composition of specific plant tissues, increase plant resistance against nematode invasion, which may deter pathogenic attacks (Mendes *et al.*, 2013). The aromatic ring of phenols, along with –OH groups, play a major role in plant defense mechanisms (Zhang *et al.*, 2018), (Guajardo-Flores *et al.*, 2013; Ramírez-Jiménez *et al.*, 2014; Wang *et al.*, 2015).

Other researchers also observed that the infection of *Meloidogyne spp* can be blocked by phenolic compounds (Brenes *et al.*, 2011). The results from the present study are consistent with the findings of Lops et al stated for *Meloidogyne* spp, the causal nematode agent of rot Knot on several horticultural crops and confirmed the antinematode effects of OMWW (LOPS *et al.*, 2008). The second finding which was recorded in the present study was the significant decrease of e *Meloidogyn spp* population, when infected tomato plans were watched with OMWW at 50% and 75%.The positive influence of the phenols on nematode growth was reported by Bevilacqua (Bevilacqua *et al.*, 2017). Martínková that new promising wild-type producers have arisen from phenolic degradation enzymes in model mixtures of actual wastewaters (Martínková *et al.*, 2016).

CONCLUSION

OMWW represents a hazardous environmental problem. In this study we discovered a new opportunity to use OMWW to control root knot nematodes under north Sinai conditions. For the development of a circular economy, which is supposed to drive the potential development of more sustainable agriculture, the valorization of waste materials is important. This study addressed the use of different concentration of OMWW in control of plant pathogenic nematodes. Tomato plants irrigated with high concentrations of OMWW is lower than those irrigated with lower concentrations.

Significance Statement

This research revealed the beneficial and important benefits of using various OMWW concentrations as an organic control measure that could be applied to an integrated root-knot nematode disease management program for tomato crops. Also this work findings are novel and opens new technology by using OMWW as a control measure that could help decrease the dependence on chemical nematicide and thus avoidits environmental hazards and also avoid the probability of developing resistance to those nematicides. It can be concluded that OMWW may provide an alternative nematicide against the root-knot nematodes.

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