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Use of Entomopathogenic Nematodes of the Genus Oscheius as Insecticides Against the Potato Tuber Moth Phthorimaea Operculella (Lepidoptera: Gelechiidae)

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ABSTRACT: Worldwide, the potato tuber moth which is the most significant potato pest considerable important challenge that is facing potato productivity and profitability in the world. At present, there are many studies demonstrating that Oscheius can control various pests and play a key role as biocontrol agents. This study was carried out at the College of Science – University of Wasit in Iraq from September 2023 to March 2024 to assessment of the effectiveness of different concentrations of the insect pathogenic nematode of the genus Oscheius against different stages of potato tuber moth under laboratory conditions. The experiment process involved more than one test on Phthorimaea operculella larvae and pupae that focused on evaluating the efficacy of two nematodes; the two Oscheius nematode species (O. myriophilus and O. tipulae) and Heterohabditis bacteriophora which is considered the commercial and local) isolates. The current study has promising results since it had good findings that the local and commercial isolates of the nematode had the potential to control the larvae of insects. The obtained data from this study reported that the local isolate was more effective in controlling insect larvae and pupae than the commercial isolate. This study recommended that important to test the biological control agents' effectiveness (fungi and nematodes) in controlling other stages of insects, adults, and eggs.

Keywords: Entomopathogenic nematodes, Oscheius, potato tuber moth, Phthorimaea operculella, Lepidoptera Gelechiidae, O. myriophilus, O. tipulae



1. INTRODUCTION

Worldwide, Potato (*Solanum tuberosum*) is the most significant vegetable crop used for feeding animals and human consumption[1]. Rice, wheat, and potato are some of the top food crops in the world, by productivity making potato crop the third-ranking[2]. Unfortunately, the potato tuber moth (PTM) (*Phthorimaea operculella*) which is the most significant potato pest considerable important challenge that is facing potato productivity and profitability in the world. In South America was originated the potato tuber moth specifically in the Andean regions[3]. The term tobacco splitworm refers to PTM which is distributed in most regions and countries of the world as a ubiquitous pest[4]. Economically, feeding of PTM larval, it's boring tunnels, and excreta deposited in tubers that occur under conditions of storage to potato tubers contribute to the most damage, causing unfit the tuber for either consumption at the local level and/or exportation[5]. Tunneling and feeding are attributed to damage resulting in the growth of bacteria or infestation of fungi[6]. The damage of potato tubers rapidly increases when the development of several generations during a period of storage. The larvae can cause damage to crops of potatoes either in the stores or in the field[7]. There are promising results in using Entomopathogenic nematodes (EPNs) in biological control as an alternative measure to many controls of pests in soil. Many studies demonstrate that the Oscheius which is the EPN genus contributes to biological control[8]. Entomopathogenic nematodes genus *Oscheius* are nematodes of soil-dwelling from a family of the *Rhabditidae*. Many

of the *Oscheius* genus species are considered nematodes of bacteriophagous free-living or scavengers. While other species of this genus are capable of killing insects. About 30 species in the *Oscheius* genus currently are recognized, and more than thirteen of these genus can kill insects[9]. Preliminary studies and experiments revealed that under laboratory conditions, nematodes including Oscheius spp. infect and kill two species of insects; *Tenebrio molitor* and *Pieris rapae*. This makes potentially the nematodes and their species vital agents for biological control. Furthermore, in China more specifically Chongming Island, *Oscheius microvilli* showed a high effectiveness level[10]. In light of this review, the aim is to summarize the important of use the entomopathogenic nematodes genus of *Oscheius* as pesticides against potato tuber moth by to assessment of the effectiveness of different concentrations of the insect pathogenic nematode of the genus *Oscheius* against different stages of potato tuber moth under laboratory conditions.

2. MATERIALS AND METHODS

Source of Galleria mellonella

Galleria mellonella were obtained from Entomology laboratory of Department of Agricultural Research - Department of Biological Control laboratories- the Ministry of Science and Technology, in Baghdad governorate.

Rearing of Galleria mellonella

The grater wax moth larva Galleria mellonella L. (Lepidoptera: Galleridae) was used for nematode baiting and to produce progeny of nematode isolates and used as a nutritional medium. The eggs of the *Galleria mellonella* insect were collected from the laboratories of the Agricultural Research Department in the Ministry of Science and Technology and placed inside plastic pots containing an artificial medium (nutritional medium) consisting of: 810 gm of jareesh, 120 g glycerin, 60 gm molasses, 10 g yeast. Under appropriate conditions, including temperature, ventilation, and lighting, at a rate of 12 hours of light and 12 hours of darkness during one day, the eggs hatched and turned into larvae within 5-10 days, and in the last instar, larvae were collected and used in nematode rearing (in the experiments). Some of these larvae were left in the plastic containers to pupate. Also, the culture of *Galleria mellonella* L. insect was reared in plastic containers with 1,500 ml volume (11 cm diameter and 15 cm height) at 25-28 °C on an artificial medium. These containers were closed with a metal screen and filter paper. Females laid the eggs on the filter paper and then collected and transferred into fresh medium. The eggs hatched within 5-10 days and the Larvae were weekly fed. After 5-6 weeks, larvae reached the last instar and were collected to be used in the experiments. Later two weeks, the female adult emerged and laid eggs

Source of Phthorimaea operculella

Phthorimaea operculella culture was obtained from infested potatoes available at Entomology laboratory of Department of Agricultural Research - Department of Biological Control laboratories- the Ministry of Science and Technology, in Baghdad governorate.

Rearing of Phthorimaea operculella

In this study, Potato tuber moth adults were collected and placed inside plastic pots. The adults were transferred to one-liter size plastic jars covered with a white muslin cloth and filter paper instead of the lid which is an oviposition preferred substrate and provided with a 10% sugar solution as a food source, while pricked potato tubers were provided for the larvae. The adult females laid eggs on a white muslin cloth. The laboratory environmental conditions were maintained at 20±25C and 60-70% relative humidity (R.H.) and photoperiod (12:12 light: dark). Daily collected the eggs and transferred them to similar jars that contained several potato tubers for the establishment of larvae under similar conditions in the laboratory. Pupae were kept and maintained in separate containers for the emergence of adults

Source of Entomopathogenic Nematodes

Local and commercial nematode were obtained from Entomology laboratory of Department of Agricultural Research - Department of Biological Control laboratories- the Ministry of Science and Technology, in Baghdad governorate.

Rearing of Entomopathogenic Nematodes

In the experiment, Petri dishes (diameter 5 cm) filled with filter paper discs were used to carry the Galleria mellonella in continuous contact with the two *Oscheius* nematode species (*O. myriophilus* and *O. tipulae*). The larvae of the Galleria mellonella insect were used as a medium for feeding and reproducing the nematodes. The rearing process began by placing the *Galleria mellonella* larvae using forceps (five larvae per dish), and these dishes were placed in the incubator under 25±3°C and 90% RH which contributed to accelerating the propagation process. It took 5 days for the appearance of the infective juveniles to be observed after examining the samples. The infective juveniles stage then were used and converted into suspension via added sterile distilled water to the Petri dish that contained the infective juveniles' stage. After this step, it was withdrawn by using a pipette and transferred to a Tissue Flask with 25 milliliters size and stored in the refrigerator at a temperature ranging between 8 -10°C.

Preparation nematode suspension

The commercial and local types of two *Oscheius* nematode species (*O. myriophilus and O. tipulae*) were used to identify the efficiency of these types on *Phthorimaea operculella*. The suspension preparation that contains nematode infective juvenile stage (IJs) was initially prepared by using a fine pipette to take 100 microliters volume of the suspension and

count the infective juvenile stages. The value of commercial and local types was of 92 IJS and 85 IJS respectively. To calculate the concentration of IJS for a 25 milliliters volume: c1/v1=c2/v2. Where:

C1	The IJS concentration in 25 milliliters of nematode suspension			
V1	The volume of Tissue Flask (25 milliliters).			
C2	The IJS concentration in 100 microliters			
V2	The volume of nematode suspension that was withdrawn with the micropipette.			

Table 1: Concentrations of ijs and sizes along with the larvae and pupae numbers that were adopted in the experiments for commercial *Oscheius* nematode species.

Sample type	Concentrations of nematode suspension infective juvenile stage/microl	Size of nematode suspension/micol		
Controls	-	-		
	_25	360		
The larvae	50	180		
The larvae	75	120		
	100	91		
	25	360		
Dungs	50	180		
Pupae	75	120		
	100	91		

Table 2: Concentrations of ijs and sizes along with the larvae and pupae numbers that were adopted in the experiments for local *O. myriophilus* nematode.

Sample type	Concentrations of nematode suspension infective juvenile stage/microl	Size of nematode suspension/microl	
Controls	-	-	
The	25	188	
larvae	50	94	
	75	62	
	100	47	
Pupae	25	188	
	50	94	
	75	62	
	100	47	

Table 3: Concentrations of ijs and sizes along with the larvae and pupae numbers that were adopted in the experiments for local *O.tipulae* nematode.

Sample type	Concentrations of nematode suspension infective juvenile stage/microl	Size of nematode suspension/micol
Controls	-	-
The	25	280
larvae	50	140
	75	93
	100	70
Pupae	25	280
	50	140
	75	93
	100	70

Statistical analysis

The Microsoft Excel 2019 program was used to enter the data. Also, the GenStat program (version 3) was used in the process of statistical analysis to test all laboratory experiment results. A completely randomized design (CRD) was used for all experiments. The rates of killing were corrected using Abbott's correction (Abbott, 1925). The statistical analysis of compatibility and interaction between the different concentration parameters for nematodes and fungi was done by

using a multifactor ANOVA design and the Fisher LSD test was used to compare the total rates with a confidence probability of 95% ($P \le 0.05$).

3. RESULTS AND DISCUSSION

The *Phthorimaea operculella Zeller* is a potato tuberworm called *Lepidoptera: Gelechiidae* which is considered the most common pests in the production area of potatoes. Pest control is difficult and extensive insecticides over the years have been used[11]. Thus, this study was designed to evaluate the performance of biological control agents in controlling *Phthorimaea operculella* growth and reproduction, which is considered an economically harmful pest, as was indicated in the first chapter. To reach acceptable results, the researcher was used different models and samples. To achieve more accurate results in calculating the insect pest mortality rates during specific periods using specific concentrations of biological control agents. So, the researcher chose a specific number of models (ten larvae and pupae).

Effect of commercial nematodes on larvae and pupae of Potato tuber moth

In this study, the larvae and the pupae of *Phthorimaea operculella* in the pre-pupation stage were taken and they were treated with commercial nematodes under appropriate laboratory conditions (temperature, humidity, and the period of lighting for the insect). The infective stages (Ijs) that are characterized by high activity and specific concentrations were taken and recorded results of seven days after the experiment was conducted by using microscopic examination for *Phthorimaea operculella* larvae and pupae that had not turned into adults.

Table (4) shows that the commercial nematode had a clear effect on the *Phthorimaea operculella* larvae and pupae stages and that the effect of suspension of the nematode increased with increasing concentration. The obtained results showed that the larval stage was more sensitive than the pupal stage represented by the highest mortality percentage recorded for treated larvae was 46.66% after 7 days, while the highest mortality percentage for treated pupae was 36.66% after 7 days at a concentration of 100 infective stages/ml.

Conc. of ijs, ijs/ ml	Mortality percentages (%)		
	The larvae	Pupae	Rate
25	16.66	10	13.33
50	30	1666	23.33
75	36.66	23.33	29.99
100	46.66	36.66	41.66
(Control)	0	0	
Rate	32.495	21.66	
LSD (0.05)	11.51	11.51	

Table 4: Effect of commercial nematodes on larvae and pupae of Potato tuber moth.

It was observed that two days after the treatment with nematodes, the larvae treated turned into pupae. The results also demonstrated that all concentrations used in experimental achieved a good mortality percentage, but with varying percentages ranging from 16.66% to 46.66% in the case of treating larvae with commercial nematodes and between 10% to 36.66% in the case of treating pupae with commercial nematodes. For treated pupae and larvae, the rate of killing was less than 50% recorded by commercial nematodes. In this study, daily monitoring of the models over seven continuous days to record any observed changes in the larvae and pupae treated with commercial nematodes. Additionally, monitoring the nematodes by microscope and observing their spread in the dishes throughout the experiment. After 7 days of treatment, observed that some of the larvae and pupae in these dishes were turned into adults, and recorded their numbers. The rest of the larvae and pupae that had incomplete development were examined and these were dissected under a microscope in order to ensure they were infected and killed by nematodes.

Concerning the rate of kill of *Phthorimaea operculella* larvae that were treated with commercial nematodes, a 16.66% killing rate was recorded for larvae with commercial nematodes at 25 infective stages/ml of concentration after 7 days. This means, that 5 out of 30 larvae treated with the nematode were confirmed infected. While the rest of the larvae completed their growth and turned into adults.

Regarding the rate of kill of *Phthorimaea operculella* larvae that were treated with commercial nematodes, a 30% killing rate was recorded for larvae with commercial nematodes at 50 infective stages/ml of concentration after 7 days. This

^{*} Each number represents an average of three replicates

means, that 9 out of 30 larvae treated with the nematode were diagnosed as infected. While the rest of the larvae completed their development and growth and turned into adults without being diagnosed as infected.

In order to test the nematode's effectiveness as an agent of biological control at a concentration of 75 infectious stages/ml, it was noted that the larvae killed percentage was 36.66%, as 11 out of 30 larvae were diagnosed with an infection. The remaining larvae (19 larvae) completed their growth and turned into adults. These larvae were examined microscopely and confirmed their infected and they were killed by commercial nematodes.

The last concentration of the commercial nematode that was used to test its effectiveness in killing *Phthorimaea operculella* was 100 infectious stages/ml. It was noted that the larvae treated with commercial nematode had a 46.66% killing percentage after 7 days of treatment. This means that the 14 larvae that did not turn into adults were examined, dissected, and observed to be infected and killed by commercial nematodes under a microscope. Thus, the highest killing percentage of larvae recorded by the highest commercial nematode concentration when treated with it.

Regarding the experiment results of treatment pupae of the *Phthorimaea operculella*, they were also treated with the same previous concentration, a 10% killing rate was recorded for pupae with commercial nematodes at 25 infective stages/ml of concentration after 7 days. This means, that 3 out of 30 pupae treated with the nematode were diagnosed as infected that present in 3 cases (each dish contain 10 pupae). These were infected after examining and dissecting under microscope, where it was noted that they were infected and killed by the nematodes spreading in them.

The second concentration that was used to test the effectiveness of commercial nematodes against the pupae of the Phthorimaea operculella was 50 infectious stages/ml. The microscopic examination and autopsy were performed on the untransformed pupae, it was noted that 5 pupae were infected and killed by nematodes, which means that the killing percentage was 16.66%.

When testing the effectiveness of commercial nematodes with a third concentration of 75 infective stages/ml on pupa, the killing percentage of pupae was 36.66% after 7 days. This means that 7 out of 30 pupae were recorded as being infected.

The last concentration that was used to test the effectiveness of commercial nematodes on pupae was 100 infective stages/ml. The killing percentage of pupae was 45.66% after 7 days, meaning that 11 out of 30 pupae were recorded as being infected. This is considered the highest killing percentage of pupae recorded by the highest concentration of the commercial nematode. The killing percentage of *Phthorimaea operculella* larvae and pupae that were treated with the commercial nematode did not reach 50%, as it was less than that for both the stage of larval and pupae.

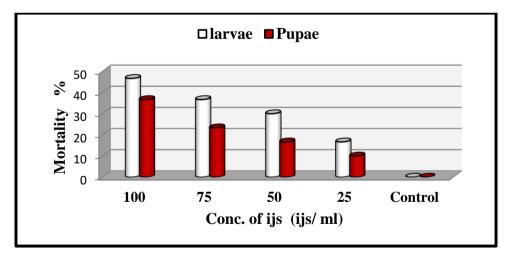


Figure 1: Percentage of larval and pupal mortality of treatment with commercial nematodes

The results indicate that increases in infecting the treated larvae and pupae due to increases in the nematode's concentration resulted in increases in the killing percentage. This means increases in the efficiency of biological control. This is consistent with many studies that have indicated that the effect of nematodes on insects increases with an increase in the nematode suspension concentration[12]. It was also noted from the results that larvae are more sensitive than pupae to the insect-pathogenic nematode. The obtained results from this study are in full agreement with IBRAHIM,A.A. et al., 2019, who concluded that that the *Ph. operculella* larvae was more susceptible than pupae to fungal infection[12]. Similarly, the results of this study align well with other research done by Heba A. A. and Gamila A. M. which showed

that *H. bacteriophora* as a biocontrol agent, has a greater effect on the larval than the pupae of the potato tuber moth since it recorded a high reduction percentages in the host insect population[13]. Aydemir Barış et al. (2023) indicated that *H. bacteriophora* was the most effective against the last instar of PTM larvae, so it had a good potential to control the larvae of PTM(14]. In Russia, The nematode of the genus *Heterorhabditis* plays a significant role in P. operculella control and it has promising results[15]. The obtained data of this study agree with IBRAHIM,A.A et al., (2019) who reported a high mortality of Ph. Operculella larvae and pupae treated with *B. bassiana* at all concentrations[12]. Entomopathogenic nematodes (EPNs) and bacteria that symbiont with it act in tandem on pests of insects and cause mortality rapidly in the pest population[16].

Effect of local nematodes (O.myriophilus and O. tipulae) on larvae and pupae of Potato tuber moth

In this study, the effect of the local nematode (*O.myriophilus* and *O. tipulae*) on *Phthorimaea operculella* larvae and pupae showed that the *O.myriophilus* nematode affected both stages and achieved a mortality rate that ranged from 26.66% to 63.33%, and the *O. tipulae* nematode affected both stages and achieved a mortality rate that ranged from 30% to 70%. As well as this effect was proportional to the concentration used in the experimental, as it increased with the increase in the concentration.

The results also showed that the highest rate of mortality achieved in the case of treating larvae with *O.myriophilus* nematode was 63.33%, and the highest mortality rate in the case of treating pupae was 60% at a concentration of 100 infective stages/ml. The highest rate of mortality achieved in the case of treating larvae with *O. tipulae* nematode was 70%, and the highest mortality rate in the case of treating pupae was 66.66% at a concentration of 100 infective stages/ml.

Conc. of	Mortality percentages (%)					
ijs, ^{ijs/ ml}	The larvae		Pupae		Rate	
	O.myriophilus	O. tipulae	O.myriophilus	O. tipulae	O.myriophilus	O. tipulae
25	26.66	30	13.33	20	28.33	16.66
50	40	43.33	36.66	40	41.66	41.66
75	53.33	56.66	50	53.33	54.99	54.99
100	63.33	70	60	66.66	64.99	66.66
Control	(0)					
Rate	45.83	49.99	39.99	41.66		
LSD	Conc		Conc		Nematode	Interaction
(0.05)	18.1	3	18.13		N.S	N. S

Table 5: Effect of local nematodes on larvae and pupae of Potato tuber moth.

Concerning the rate of kill of *Phthorimaea operculella* larvae that were treated with *O.myriophilus and O. tipulae* nematode, a 26.66% and 30% killing rate was recorded for larvae with O.myriophilus and O. tipulae nematode respectively at 25 infective stages/ml of concentration after 7 days. This means, that 8 and 9 out of 30 larvae treated with the nematode were confirmed infected. While the rest of the larvae completed their growth and turned into adults.

Regarding the rate of kill of *Phthorimaea operculella* larvae that were treated with *O.myriophilus and O. tipulae* nematode, a 40% and 43.33% killing rate was recorded for larvae with O *O.myriophilus and O. tipulae* nematode respectively at 50 infective stages/ml of concentration after 7 days. This means, that 12 and 13 out of 30 larvae treated with the nematode were diagnosed as infected. While the rest of the larvae completed their development and growth and turned into adults without being diagnosed as infected.

In order to test the *O.myriophilus and O. tipulae* nematode's effectiveness as an agent of biological control at a concentration of 75 infectious stages/ml, it was noted that the larvae killed percentage was 53.33% and 56.66% for larvae with *O.myriophilus and O. tipulae* nematode respectively, as 16 and 17 out of 30 larvae were diagnosed with an infection. The remaining larvae (14 and 13 larvae) completed their growth and turned into adults. These larvae were examined microscopely and confirmed their infected and they were killed by commercial nematodes.

The last concentration of the *O.myriophilus and O. tipulae* nematode nematode that was used to test its effectiveness in killing *Phthorimaea operculella* was 100 infectious stages/ml. It was noted that the larvae treated with O *O.myriophilus and O. tipulae* nematode had a 63.33% and 70% respectively of killing percentage after 7 days of treatment. This means that the 14 and 13 larvae that did not turn into adults were examined, dissected, and observed to be infected and killed by

commercial nematodes under a microscope. Thus, the highest killing percentage of larvae recorded by the highest local nematode concentration when treated with it.

Regarding the experiment results of treatment pupae of the *Phthorimaea operculella*, they were also treated with the same previous concentration, a 13.33% and 20% killing rate was recorded for pupae with *O.myriophilus and O. tipulae* nematodes respectively at 25 infective stages/ml of concentration after 7 days. This means, that 4 and 6 out of 30 pupae treated with the nematode were diagnosed as infected that present in 3 cases (each dish contain 10 pupae). These were infected after examining and dissecting under microscope, where it was noted that they were infected and killed by the nematodes spreading in them.

The second concentration that was used to test the effectiveness of *O.myriophilus and O. tipulae* nematodes against the pupae of the *Phthorimaea operculella* was 50 infectious stages/ml. The microscopic examination and autopsy were performed on the untransformed pupae, it was noted that 11 and 12 pupae were infected and killed by *O.myriophilus and O. tipulae* nematodes, which means that the killing percentage was 36.66% and 40% respectively.

When testing the effectiveness of commercial nematodes with a third concentration of 75 infective stages/ml on pupa, the killing percentage of pupae with *O.myriophilus and O. tipulae* nematodes was 50% and 53.33% respectively after 7 days. This means that 15 and 16 out of 30 pupae were recorded as being infected.

The last concentration that was used to test the effectiveness of *O.myriophilus and O. tipulae* nematodes nematodes on pupae was 100 infective stages/ml. The killing percentage of pupae was 66.66% and 64.99% respectively after 7 days, meaning that 18 and 20 out of 30 pupae were recorded as being infected. This is considered the highest killing percentage of pupae recorded by the highest concentration of the local nematode. The killing percentage of *Phthorimaea operculella* larvae and pupae that were treated with the local nematode more reach 50%, as it was more than that for both the stage of larval and pupae. The results indicate that increases in infecting the treated larvae and pupae due to increases in the local nematode's concentration resulted in increases in the killing percentage. This means increases in the efficiency of biological control.

From the above, the highest percentage of deaths recorded by the highest concentration of local and commercial nematodes was 100 Infectious stage/ml. This indicates that the effect is proportional to the concentration. It was also found that the larvae-killing percentage is higher than the pupae-killing percentage for all concentrations and in both types of nematodes. This may be attributed to the fact that the pupae are surrounded by a cocoon that protects the pupae from external influences to preserve the growth of insects inside it and this thing is not present in the larvae, as they were directly exposed to the biological control agent (local and commercial nematode) and the nematodes of both types were able to reach the larvae and penetrate them easily before they became impossible, and within two days of treatment with the local and commercial nematode, and this gave high killing percentages as compared to the pupae mortality percentages.

Additionally, to the fact that the larvae contain respiratory openings, which are considered suitable for entering the infective stages of the nematode. Although the nematodes were able to penetrate the cocoon and infect the developing insect (the pupa), achieving a rate of killing that may be small compared to the killing rate of larvae treated with commercial and local nematodes, it is somewhat acceptable (Figure 2, 3). Several Oscheius species have been reported to reproduce in insects and/or kill them such as *O.myriophilus and O. tipulae* [17]. *Oscheius* nematodes are significant organisms for the evolutionary dynamics of symbiotical relationships with populations of bacteria and unraveling the intricacies of pathogenicity entomopathogenic nematode[18]. The current study findings are also in agreement with those obtained from another study done by Ameni Loulou, who demonstrated that the rapid capacity of insect-killing by *O. tipulae* nematodes[9].

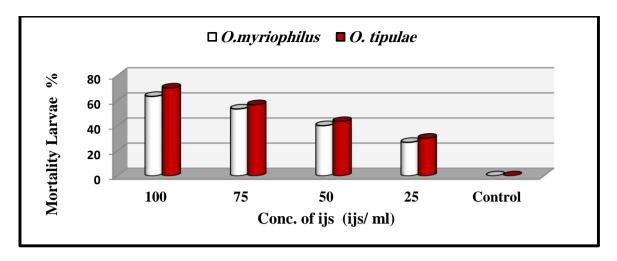


Figure 2: Percentage of larval mortality of treatment with local nematodes

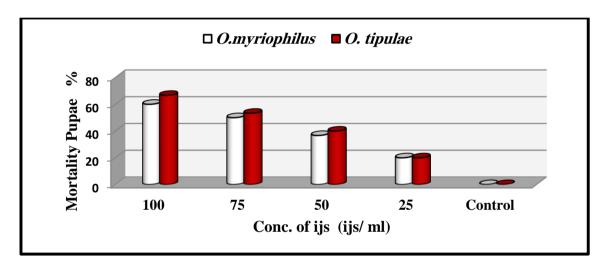


Figure 3: Percentage of pupal mortality of treatment with local nematodes

In comparison between the results obtained from the treatment of the larvae and pupae with the local and commercial nematode as a biological control agent, it noted that the percentages of killing recorded by the local nematode were higher than those recorded by the commercial nematode. This can be attributed to the local nematode's adaptation to external conditions and its ability to penetrate the treated insect and infest its stages (larval and pupal) with a higher efficiency than that of the commercial nematode as appearance clearly by microscopic examination which found the infestation was severe and a large density of growth and reproduction inside the insect as compare with the commercial type.

It is clear from the results of this study that the nematode is an effective biological control agent against the larvae and pupae of the *Phthorimaea operculella* after providing appropriate conditions for its growth. This is consistent with what was mentioned in the studies. Ahmed A. A. et al. (2021) reported that nematodes are fatal agents of biological control that treat a wide range of significant insect pests economically[19].

Conclusions: Nematodes as a biocontrol agent, have a higher efficiency on the larval than the pupae of the potato tuber moth since it recorded high reduction percentages in the host insect population. These nematodes' effectiveness increases with an increase in the nematode suspension concentration. It was also noted from the results that commercial nematodes are less virulent than local nematodes against the pupae and larvae of the potato tuber moth. In summary, the usage of entomopathogenic nematodes genus of *Oscheius* as pesticides against potato tuber moth is a more effective and environmentally-friendly biological control as well as it is an alternative measure for many soil pest control, cheap, locally available, easy to be prepared, and safe to be used for insect control. However, there is still a lack of biological control in terms of the use of Oscheius as a pesticide against other solanaceous plants. A field study must be conducted on P. operculella larvae and pupae to determine the biological control agents' efficiency in field conditions.

REFERENCES

- [1] Z. K. M. Abdelsalam, A. S. Ezzat, I. A. A. Tantawy, N. S. Youssef, and S. H. G. El-, "Effect of NaCl salinity stress on potato (Solanum tuberosum L.) plantlets grown and development under in vitro conditions," Sci. J. Agric. Sci., vol. 3, no. 2, pp. 1–12, 2021, doi: 10.21608/sjas.2021.84222.1125.
- [2] C. G. Kuyu, Y. B. Tola, and G. G. Abdi, "Heliyon Study on post-harvest quantitative and qualitative losses of potato tubers from two different road access districts of Jimma zone, South West Ethiopia," *Heliyon*, vol. 5, no. June, p. e02272, 2019, doi: 10.1016/j.heliyon.2019.e02272.
- J. Jung, S. Lee, K. Kim, S. Jeon, S. Jung, and W. Lee, "The Potential Distribution of the Potato Tuber Moth (Phthorimaea Operculella) Based on Climate and Host Availability of Potato †," *Agronomy*, vol. 10, no. 12, pp. 2–18, 2020, doi: 10.3390/agronomy10010012.
- [4] X. LI, X. ge ZHANG, C. XIAO, Y. lin GAO, and W. xia DONG, "Behavioral responses of potato tuber moth (Phthorimaea operculella) to tobacco plant volatiles," *J. Integr. Agric.*, vol. 19, no. 2, pp. 325–332, 2020, doi: 10.1016/S2095-3119(19)62663-8.
- [5] A.-M. Taha and R. Hassan, "Possibility for laboratory mass rearing of the potato tuber moth Phthorimaea operculella (Lepidoptera: Gelechiidae) through simplified steps and procedures," *Egypt. J. Plant Prot. Res. Inst.* (2021), vol. 4, no. 2, pp. 182–192, 2021, [Online]. Available: www.ejppri.eg.net
- [6] A. Adhikari, A. K. Shrestha, S. Timsina, and A. Adhikari, "Efficacy of biopesticides in management of potato tuber moth, Phthorimaea operculella (Zeller), in potato under storage," *J. Agric. Food Res.*, vol. 10, no. October, p. 100411, 2022, doi: 10.1016/j.jafr.2022.100411.
- [7] P. S. Prasad, H. Amarananjundeswara, S. Shetty, and G. C. Sandhya, "Management of Potato Tuber Moth during Storage by using Biological Agents in Hassan District of Karnataka," *Int. J. Curr. Microbiol. Appl. Sci.*, vol. 9, no. 9, pp. 465–470, 2020, doi: 10.20546/ijcmas.2020.909.059.
- [8] E. Foelkela and L. B. M. and G. N. *, M. Vossb, "Isolation of entomopathogenic nematodes in an apple orchard in Southern Brazil and its virulence to Anastrepha fraterculus (Diptera: Tephritidae) larvae, under laboratory conditions," *Braz. J. Biol.*, vol. 77, no. 1, pp. 22–28, 2017, doi: 10.1590/1519-6984.08315.
- [9] A. Loulou *et al.*, "Potential of Oscheius tipulae nematodes as biological control agents against Ceratitis capitata," *PLoS One*, vol. 17, no. 6, pp. 1–19, 2022, doi: 10.1371/journal.pone.0269106.
- [10] R. Darsouei, J. Karimi, and E. Shokoohi, "Oscheius rugaoensis and Pristionchus maupasi, two new records of entomophilic nematodes from Iran," *Russ. J. Nematol.*, vol. 22, no. 2, pp. 141–155, 2014.
- [11] G. Y. YUAN Hui-guo, WU Sheng-yong, LEI Zhong-ren, Silvia I. Rondon, "Sub-lethal effects of Beauveria bassiana (Balsamo) on field populations of the potato tuberworm Phthorimaea operculella Zeller in China," *J. Integr. Agric.*, vol. 17, no. 4, pp. 911–918, 2018.
- [12] S. A. M. IBRAHIM, A.A., SANEYA R.M.FARAG, "COMBINED EFFECT OF BEAUVERIA BASSIANA (BALS.) AND GAMMA IRRADIATION ON POTATO TUBER MOTH PHTHORIMAEA OPERCULELLA (ZELLER)," *Egypt. J. Agric. Res.*, vol. 97, no. 3, pp. 559–570, 2019.
- [13] H. A. A. Al-Ghnam and G. A. M. Heikal, "Evaluating the Role of Entomopathogenic Nematodes for the Biological Control of the Potato Tuber Moth, Phthorimaeao perculella under Laboratory Conditions.," *J. Plant Prot. Path.*, vol. 8, no. 11, pp. 577–580, 2017.
- [14] A. Barış and H. Ç. and C. Y., Mürşide Yağcı, "Efficacy of a native entomopathogenic nematode Heterorhabditis bacteriophora (isolate Z-1) against potato tuber moth (Phthorimaea operculella (Zeller) (Lepidoptera: Gelechiidae) in Turkey," *Egypt. J. Biol. Pest Control*, vol. 33, no. 35, pp. 2–7, 2023.
- [15] S. I. R. and Y. Gao, *Moths Pests of Potato*, *Maize and Sugar Beet*. 2018. doi: http://dx.doi.org/10.5772/intechopen.81934.
- [16] K. R. S. Manochaya, Shashikant Udikeri, B.S. Srinath, Mantri Sairam, Srinivas V. Bandlamori, "In vivo culturing of entomopathogenic nematodes for biological control of insect pests: A review," *J. Nat. Pestic. Res.*, vol. 1, no. 100005 Many, pp. 1–8, 2022, doi: https://doi.org/10.1016/j.napere.2022.100005.
- [17] S. E. Karimi J, Rezaei N, "ADDITION OF A NEW INSECT PARASITIC NEMATODE, OSCHEIUS TIPULAE, TO IRANIAN FAUNA," *a. Nematropica*, vol. 48, pp. 45–54, 2018.
- [18] K. H. Taiki Sugiyama, Daiki Sawanomukai, Seiya Nagae, "Exploring the pathogenicity of Oscheius sp. KHA501 and its potential in association with entomopathogenic bacteria," *Nematol. Res.*, vol. 53, no. 1/2, pp. 1–10, 2023.
- [19] A. E. E. and A. E. A. E. Ahmed A. A. Aioub , Ramadan M. El-Ashry, Ahmed S. Hashem, "Compatibility of entomopathogenic nematodes with insecticides against the cabbage white butterfly, Pieris rapae L. (Lepidoptera: Pieridae)," *Egypt. J. Biol. Pest Control*, vol. 31, no. 153, pp. 2–12, 2021, doi: https://doi.org/10.1186/s41938-021-00498-z.
- [1] Z. K. M. Abdelsalam, A. S. Ezzat, I. A. A. Tantawy, N. S. Youssef, and S. H. G. El-, "Effect of NaCl salinity stress on potato (Solanum tuberosum L.) plantlets grown and development under in vitro conditions," *Sci. J. Agric. Sci.*, vol. 3, no. 2, pp. 1–12, 2021, doi: 10.21608/sjas.2021.84222.1125.
- [2] C. G. Kuyu, Y. B. Tola, and G. G. Abdi, "Heliyon Study on post-harvest quantitative and qualitative losses of potato tubers from two different road access districts of Jimma zone, South West Ethiopia," *Heliyon*, vol. 5, no.

- June, p. e02272, 2019, doi: 10.1016/j.heliyon.2019.e02272.
- J. Jung, S. Lee, K. Kim, S. Jeon, S. Jung, and W. Lee, "The Potential Distribution of the Potato Tuber Moth (Phthorimaea Operculella) Based on Climate and Host Availability of Potato †," *Agronomy*, vol. 10, no. 12, pp. 2–18, 2020, doi: 10.3390/agronomy10010012.
- [4] X. LI, X. ge ZHANG, C. XIAO, Y. lin GAO, and W. xia DONG, "Behavioral responses of potato tuber moth (Phthorimaea operculella) to tobacco plant volatiles," *J. Integr. Agric.*, vol. 19, no. 2, pp. 325–332, 2020, doi: 10.1016/S2095-3119(19)62663-8.
- [5] A.-M. Taha and R. Hassan, "Possibility for laboratory mass rearing of the potato tuber moth Phthorimaea operculella (Lepidoptera: Gelechiidae) through simplified steps and procedures," *Egypt. J. Plant Prot. Res. Inst.* (2021), vol. 4, no. 2, pp. 182–192, 2021, [Online]. Available: www.ejppri.eg.net
- [6] A. Adhikari, A. K. Shrestha, S. Timsina, and A. Adhikari, "Efficacy of biopesticides in management of potato tuber moth, Phthorimaea operculella (Zeller), in potato under storage," *J. Agric. Food Res.*, vol. 10, no. October, p. 100411, 2022, doi: 10.1016/j.jafr.2022.100411.
- [7] P. S. Prasad, H. Amarananjundeswara, S. Shetty, and G. C. Sandhya, "Management of Potato Tuber Moth during Storage by using Biological Agents in Hassan District of Karnataka," *Int. J. Curr. Microbiol. Appl. Sci.*, vol. 9, no. 9, pp. 465–470, 2020, doi: 10.20546/ijcmas.2020.909.059.
- [8] E. Foelkela and L. B. M. and G. N. *, M. Vossb, "Isolation of entomopathogenic nematodes in an apple orchard in Southern Brazil and its virulence to Anastrepha fraterculus (Diptera: Tephritidae) larvae, under laboratory conditions," *Braz. J. Biol.*, vol. 77, no. 1, pp. 22–28, 2017, doi: 10.1590/1519-6984.08315.
- [9] A. Loulou *et al.*, "Potential of Oscheius tipulae nematodes as biological control agents against Ceratitis capitata," *PLoS One*, vol. 17, no. 6, pp. 1–19, 2022, doi: 10.1371/journal.pone.0269106.
- [10] R. Darsouei, J. Karimi, and E. Shokoohi, "Oscheius rugaoensis and Pristionchus maupasi, two new records of entomophilic nematodes from Iran," *Russ. J. Nematol.*, vol. 22, no. 2, pp. 141–155, 2014.
- [11] G. Y. YUAN Hui-guo, WU Sheng-yong, LEI Zhong-ren, Silvia I. Rondon, "Sub-lethal effects of Beauveria bassiana (Balsamo) on field populations of the potato tuberworm Phthorimaea operculella Zeller in China," *J. Integr. Agric.*, vol. 17, no. 4, pp. 911–918, 2018.
- [12] S. A. M. IBRAHIM, A.A., SANEYA R.M.FARAG, "COMBINED EFFECT OF BEAUVERIA BASSIANA (BALS.) AND GAMMA IRRADIATION ON POTATO TUBER MOTH PHTHORIMAEA OPERCULELLA (ZELLER)," *Egypt. J. Agric. Res.*, vol. 97, no. 3, pp. 559–570, 2019.
- [13] H. A. A. Al-Ghnam and G. A. M. Heikal, "Evaluating the Role of Entomopathogenic Nematodes for the Biological Control of the Potato Tuber Moth, Phthorimaeao perculella under Laboratory Conditions.," *J. Plant Prot. Path.*, vol. 8, no. 11, pp. 577–580, 2017.
- [14] A. Barış and H. Ç. and C. Y., Mürşide Yağcı, "Efficacy of a native entomopathogenic nematode Heterorhabditis bacteriophora (isolate Z-1) against potato tuber moth (Phthorimaea operculella (Zeller) (Lepidoptera: Gelechiidae) in Turkey," *Egypt. J. Biol. Pest Control*, vol. 33, no. 35, pp. 2–7, 2023.
- [15] S. I. R. and Y. Gao, *Moths Pests of Potato*, *Maize and Sugar Beet*. 2018. doi: http://dx.doi.org/10.5772/intechopen.81934.
- [16] K. R. S. Manochaya, Shashikant Udikeri, B.S. Srinath, Mantri Sairam, Srinivas V. Bandlamori, "In vivo culturing of entomopathogenic nematodes for biological control of insect pests: A review," *J. Nat. Pestic. Res.*, vol. 1, no. 100005 Many, pp. 1–8, 2022, doi: https://doi.org/10.1016/j.napere.2022.100005.
- [17] S. E. Karimi J, Rezaei N, "ADDITION OF A NEW INSECT PARASITIC NEMATODE, OSCHEIUS TIPULAE, TO IRANIAN FAUNA," *a. Nematropica*, vol. 48, pp. 45–54, 2018.
- [18] K. H. Taiki Sugiyama, Daiki Sawanomukai, Seiya Nagae, "Exploring the pathogenicity of Oscheius sp. KHA501 and its potential in association with entomopathogenic bacteria," *Nematol. Res.*, vol. 53, no. 1/2, pp. 1–10, 2023.
- [19] A. E. E. and A. E. A. E. Ahmed A. A. Aioub , Ramadan M. El-Ashry, Ahmed S. Hashem, "Compatibility of entomopathogenic nematodes with insecticides against the cabbage white butterfly, Pieris rapae L. (Lepidoptera: Pieridae)," *Egypt. J. Biol. Pest Control*, vol. 31, no. 153, pp. 2–12, 2021, doi: https://doi.org/10.1186/s41938-021-00498-z.