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Evaluation of some microbial compounds against land snail species under field conditions at  
Kafr El-Sheikh Governorate

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Abstract

This study was conducted at Abu-Abdalla village, Sedy Salem district, Kafr El-Sheikh Governorate during two successive seasons 2021/2022. To evaluate the bactericidal activity of protecto and the fungicidal activity of biofly on population density reduction of various land snail species infesting Egyptian clover (*Trifolium alexandrinum*) as a field crop and orange (*Citrus sinensis*) trees as orchard crop, using rates (500, 250 and 125 gm /200 L.W./Fadden and (100, 75 and 50 cm/100L.W /Fadden), respectively using spray methods. Same tested compound (Protecto and Biofly) at the same rates and method of application were used against *Monacha cantiana* (Montagu) (Gastropoda: Hygromiidae) and *Theba pisana* (Müller) (Gastropoda: Helicidae) snails infesting orange (*Citrus sinensis*) trees at the same location. In addition, Protecto and Biofly were tested as poison bait in concentrations of 5, 10 and 15 % against *M. cantiana* and *T. pisana* infesting orange trees. The tested compound reduced *M. cantiana* population at the different concentrations with variable degrees. The reduction of the population started with low values after the second day of treatment, but the reduction increased gradually to reach its maximum at the end of tested period (15 days).

Introduction

In Egypt, the destructiveness of land snails is far greater today than in former time (Kassab and Daoud, 1964; El-Okda, 1983 and Abdallah *et al.*, 1992). Land snails have been concentrated in the northern Governorate of Delta region and in Upper Egypt to several host plants. These animals become active all year and their activities increase in spring and autumn seasons

(Godan, 1983). These snails become an important agricultural pest, causing great damage to different crops and orchards production in different localities.

These snails were recorded with relatively high population density on major economic crops at Kafr El-Sheikh Governorate (Sharsher *et al.*, 1996). Until now, these pests are controlled chemically by synthetic molluscicides or insecticides

(Crowell, 1967 and El-Okda,1981). These chemical compounds cause environmental contamination which gives rise to residues in food, fruits, and water.

Therefore, it was necessary to apply some methods as a biological control to the management of the snails population under field conditions by using microbial compounds (Abdel Megeed et al., 1998; Zedan et al.1999; Sleem, 2002; Idrees, 2003; Gaber et al., 2005; Khider et al ., 2006 and Mortada et al., 2009 ) .

### Materials and methods

#### Effect of two microbial compounds on various land snail species infesting different crops:

##### 1. Tested microbial compounds:

###### -Protecto

*Bacillus thuringiensis* var Kurstaki active ingredient 9.4% and inert ingredient 90.6%.

###### - Biofly

*Beauveria bassiana* 30 x 10<sup>3</sup> conidia / cm.

2. To evaluate the bactericidal activity of protecto and the fungicidal activity of biofly on population density reduction of various land snail species infesting Egyptian clover (*Trifolium alexandrinum*) as a field crop and orange (*Citrus sinensis*) trees as orchard crop, field experimental was carried out at Abu- Abdalla village, Sedy Salem district, Kafr El-Sheikh Governorate during successive season 2021 /2022. For these studies two experiments areas ( One feddan each) were chosen, the first area was cultivated with Egyptian clover (*Trifolium alexandrinum*) which high infested with *Monacha cantiana* (Montagu) (Gastropoda: Hygromiidae) and *Succinea putris* (L.) (Pulmonata: Succineidae) snails . These snails were found in clover plantations in high density compared with other snail species. The second area was cultivated with citrus trees, orange (*C. sinensis*) which were infested with *M. cantiana* and *Theba*

*pisana* (Müller) (Gastropoda: Helicidae) snails. The tested biocides were applied at rats (125, 250 and 500 gm /200 L.W/Feddan) for Protecto and (50, 70 and 100 ml/100 L.W/Feddan) for Biofly, spraying method was used. . The experiments were conducted in a completely randomized block design consisting of two treatments including control and replicated five times (5x5m each). The remaining experimental area acted as a buffer between plots. The tested compounds were sprayed after three days of irrigation on the damp soil in the evening. In the case of orange trees, five trees infested with *M.cantiana* and *T. pisana* snails for each treatment and snails population on trees trunks (One meter) and on soil surrounded trees ( One meter diameter) were counted . Protecto and Biofly were used as poison baits, the rate of 2 ,3, 5 and 10 gm % for Protecto and 2 ,3, 5, and 10 cm% for Biofly. Toxic bran baits used against snails infested orange trees, placed plastic sheets around trees (El- Okda , 1983 ). The snails population (Snails / 0.5 m<sup>2</sup>) was recorded before and after 1, 2, 3, 5, 7, 9, 12 and 15 days post treatment. Reduction percentages were calculated according to **Henderson and Tilton equation (1955):**

$$R \% = (1 - t_2 \times r_1 / t_1 \times r_2) \times 100$$

Whereas:

t = Population size at treated area

r = Population size at untreated area

t<sub>1</sub> x r<sub>1</sub> = Pre-treatment

t<sub>2</sub> x r<sub>2</sub> = post-treatment

### Results and discussion

#### Effect of certain microbial compounds against some land snails infesting different crops under field conditions:

The efficacy of Protecto and Biofly was evaluated against *M. cantiana* and *S.putris* infesting Egyptian clover (*T. alexandrinum*) at Abu- Abdalla village, using rates (500 ,250 and 125 gm /200 L.W./Fadden and (100, 75 and 50 cm/100L.W /Fadden), respectively using spray methods. The same tested

compound at the same rates and method of application was used against *M. cantiana* and *T. pisana* snails infesting orange (*C. sinensis*) trees at the same location. In addition to, Protecto and Biofly were tested as poison baits in concentrations of 5, 10 and 15 % against *M.cantiana* and *T.pisana* infesting orange trees . The tested compound reduced *M. cantiana* population at the different concentrations with variable degrees. The reduction of the population started with low values after the second day of treatment, but the reduction increased gradually to reach its maximum at the end of the tested period (15 days).

**Table (1) : Effect of various microbial compounds against *Monacha cantiana* snails clover using spray method under field conditions during spring season 2022.**

Tested compounds	Tested rates	% Reduction after days								Mean
		1	2	3	5	7	9	12	15	
Protecto	500 mg	0	32.1	49.6	60.1	79.3	84.5	89.6	93.5	61.0
	250	0	19.6	28.5	47.5	61.2	70.5	76.2	80.1	48.0
	125	0	0	16.1	38.2	63.0	66.1	69.3	71.5	40.5
Biofly	500 cm	0	19.1	33.6	57.5	60.0	64.1	70.2	83.1	48.5
	250	0	17.2	19.0	36.2	47.2	53.0	68.1	69.5	38.8
	125	0	0	13.0	21.5	29.0	33.6	49.5	61.3	26.0

Data in Table (2) showed that, Protecto at rates 500, 250 and 125 gm / 200L.W/ Feddan reduced *S. putris* population snails at 2<sup>nd</sup> and 15<sup>th</sup> days post treatment with (41.6 and 83.1%), (20.4 and 73.0 %) and (16.1 and 51.2 %) , respectively. The average reduction % for the tested rates after 15 days post treatment

**Table (2) : Effect of some microbial compounds against *Succenia putris* snails on clover using a spray method under field conditions during spring season 2022.**

Tested compound	Tested rates	%Reduction after days								Mean
		1	2	3	5	7	9	12	15	
Protecto	500 mg	0	41.6	39.0	46.3	60.9	68.2	76.3	83.1	52.3
	250	0	20.4	31.0	43.5	55.1	64.2	69.2	73.0	44.6
	125	0	16.1	19.7	31.7	42.2	44.1	47.1	51.2	32.5
Biofly	100 cm	0	23.1	43.7	64.7	67.2	74.0	78.1	80.0	53.9
	75	0	17.6	41.0	58.5	67.7	72.0	71.0	72.0	50
	50	0	15.2	27.4	49.0	54.9	60.7	70.3	61.5	42.4

Data in Table (3 ) showed that , applied Protecto at rates 500 , 250 and 125 gm/200 L.W/Feddan to reduced population snails *M.chantiana* on orange trees at 2<sup>nd</sup>

Data in Table (1) showed t respectively ,t rates 500 , 250 and 125 gm /200L.W/Feddan reduced *M. cantiana* population on clover at the 2<sup>nd</sup> and 15<sup>th</sup> day post treatment with (32.1 and 93%) , (19.6 and 80.1 %) and (16.1 and 71,6 %), respectively The average rate of reduction for the three tested rates after 15 days post treatment were 61.0 , 45.0 and 40.5 % , while Biofly at rates of 100 , 75 and 50 cm /100 L.W/Feddan exhibited (19.1 and 83.1% ) , ( 17.2 and 69.5 %) and (13.2 and 61.3 %) reduction at 2<sup>nd</sup> and 15<sup>th</sup> days post treatment respectively, with average % reduction of 48.5, 38.8 and 26.0 %.

was 52, 3, 44.6 and 31.5 % , respectively. On the other hand, Biofly at rates 100, 75 and 50 cm /100L.W/Feddan exhibited (23.1 and 80.0%), (17.6 and 72.0 %) and (15.2 and 61.5%) reductions at 2<sup>nd</sup> and 15<sup>th</sup> post treatment, respectively with average % reduction of 53.9, 50.0 and 42.4% at the same tested period and concentration.

and 15<sup>th</sup> days post treatment with ( 21.6 and 74.1% ) , ( 26.0 and 64.7% )and ( 0 and 60.6% ) , respectively .The average % reduction for the three tested concentrations

after 15 days post treatment were 42.9 , 38.3 and 30.5 % , respectively while Biofly at rates 100 ,75 and 50 cm / 100 L.W/ Feddan of orange trees were ( 18.6 and

59.6% ) , ( 23.4 and 46.3% ) and ( 0 and 30.4% ) reduction at 2<sup>nd</sup> and 15<sup>th</sup> post treatment ,respectively with average reduction% of 32.7 , 31.4 and 17.6% .

**Table (3): Effect of two microbial compounds against *Monacha cantiana* snails on orange as spray method under field conditions during spring season 2022.**

Tested compound	Tested rates	% reduction after days								Mean
		1	2	3	5	7	9	12	15	
Protecto	500 mg	0	21.6	34.3	39.3	46.1	59.3	68.2	74.1	42.9
	250	0	26.0	31.0	33.6	49.1	50.6	51.1	64.7	38.3
	125	0	0	20.3	35.1	38.2	40.4	49.2	60.5	30.5
Biofly	100 cm	0	18.6	24.3	22.7	36.0	49.2	51.3	59.6	32.7
	75	0	23.4	29.4	26.3	30.0	48.4	45.2	46.3	31.4
	50	0	0	18.7	20.0	20.1	23.2	28.1	30.4	17.6

Data in Table ( 4 ) showed that application of Protecto at the same rates reduced *T.pisana* snails population at 2<sup>nd</sup> and 15<sup>th</sup> days post treatment with ( 39.2 and 81.3% ) , (27.1 and 69.4% ) and 18.3 and 59.7 % )respectively with average %reduction 50.3 , 41.4 and 33.2% , while Biofly at the same

rates on orange trees exhibited ( 24.1 and 70.3% ) , (16.3 and 64.1% ) and ( 0 and 51.5% ) .The following results showed that effect of various bio-compounds as poison baits against *M.chantiana* and *T.pisana* snails infesting orange trees at the same location during Spring season of 2022.

**Table (4): Effect of two microbial compounds against *Theba pisana* snails on orange trees as a spray method under field conditions during spring season 2022.**

Tested compounds	Tested rates	%Reduction after days								Mean
		1	2	3	5	7	9	12	15	
Protecto	500 mg	0	39.2	43.5	50.1	50.0	62.1	76.1	81.3	50.3
	250	0	27.1	30.2	39.4	46.8	57.2	61.3	69.4	41.4
	125	0	18.3	29.1	31.1	30.2	46.1	51.3	59.7	33.2
Biofly	100 cm	0	24.1	39.3	48.0	59.0	68.0	70.1	70.3	47.4
	75	0	16.3	39.34.6	33.5	39.0	47.1	64.1	64.1	41.4
	50	0	0	11.0	29.1	40.3	42.0	51.5	51.5	28.0

Data in Table (5) showed that, the biocid Protecto at rates 15, 10 and 5 gm % reduced *M. chantiana* snails population at 2<sup>nd</sup> and 15<sup>th</sup> days post treatment with (21.6 and 81.3%), ( 14.0 and 69.7% ) and ( 0 and 48.1% ) , respectively . The average % reduction for

the three rates after 15 days post treatment was 41.4, 30.7 and 25.2%, respectively. Biofly exhibited (11.2 and 64.1%), (0 and 60%) and (0 and 28.1%) with average 29 .9 ,27.8 and 15.3%, respectively at the same tested period and concentration.

**Table (5): Effect of two microbial compounds against *Monacha cantiana* snails on orange trees as a poison bait under field conditions during spring season 2022.**

Tested compounds	Tested rates	%Reduction after days								Mean
		1	2	3	5	7	9	12	15	
Protecto	15 gm	0	21.6	29.3	33.2	41.6	58.1	66.1	81.3	41.4
	10	0	14.0	21.2	24.0	29.6	36.1	51.2	69.7	30.7
	5	0	0	16.1	30.0	30.6	33.1	40.6	48.1	25.2
Biofly	15	0	11.2	19.3	22.0	27.6	42.7	51.9	64.1	29.9
	10	0	0	16.3	24.3	33.4	39.6	48.7	60.0	27.8
	5	0	0	12.0	18.0	20.0	20.3	24.0	28.1	15.3

Data in Table (6) showed that, Protecto at rates 15, 10 and 5 gm % reduced *T. pisana* snails population at 2<sup>nd</sup> and 15<sup>th</sup> days post treatment with ( 31.6 and 70.2% ) ,(19.6 and 61.3% ) and ( 0 and 49.0% ), respectively with average 41.6 , 37.2 and

27.0% , respectively at the same tested period and concentrations. Also, Biofly exhibited (10.1 and 60.0%) , ( 0 and 49,0% ) and ( 0 and 40%) with average % reduction 25.3 , 22,5 and 20.5% respectively.

**Table (6): Effect of two microbial compounds against *Theba pisana* snails on orange trees as a poison bait under field conditions during spring season 2022.**

Tested compounds	Tested rates	% Reduction after days								Mean
		1	2	3	5	7	9	12	15	
Protecto	15 gm	0	31.6	39.2	36.8	48.1	49.0	58.0	70.2	41.6
	10	0	19.6	30.1	38.5	38.0	53.1	57.1	61.3	37.2
	5	0	0	16.3	28.4	41.6	37.5	42.6	49.0	27.0
Biofly	15	0	10.1	19.3	18.3	24.0	29/0	41.9	60.0	25.3
	10	0	0	14.6	21.6	26.0	28.0	40.5	49.0	22.5
	5	0	0	9.0	15.1	24.6	39.1	36.3	40.0	20.5

These results agree with those, Lokma and Al-Harby (1999) tested Protecto 9.4% containing *B. thuringiensis* as a biocid under laboratory and field conditions against the snails *Monacha cartusiana* (Muller) (Gastropoda:Helicoidea) and *Rumina decollate* (L.) (Gastropoda: Subulinidae). The results revealed that Protecto as a soking spray was more effective than baits against the two snails under laboratory conditions. Soking spray method gave high mortality percentage, reaching 100% for *M. cartusiana* when used at concentration of 10 gm of protecto 9.4% / 100 ml water, while mortality percentage did not exceed 56% for *R. decollate*. At the same time, toxic baits application used at 10 gm of Protecto 9.4% /100gm bran gave 90% mortality against *M. cartusiana* and 52% against *R. decollate*. The field experimental trial indicated that mortality was caused by using Protecto 9.4%at 10 gm /100 ml water concentration as soking spray against *Monacha* sp. Was 80%. The obtained results encourage more studies for estimating the potential of biocides against land snails. Sleem (2002) studied that ,the effect of vertemic on the digestive gland of the land snails *E. vermiculata* , he found that a low sublethal dose of vertemic (0.05 mg of Abamactine /snail / day ) induced tubule cells

vaculation of digestive gland and on increase in the yellowish spherules , while higher sublethal dose of vertemic ( 0.15 mgob Abamactin /snail/day ) caused sever general damage in the digestive tubules , tubular epithelial degeneration and marked loss of the entire digestive gland structure , widening of tubular and complete necrosis.

Idrees (2003) tested both *B. thuringiensis* var *Barlliner* and *B. thuringiensis* var *Karstaki* in addition to *B. bassiana* 3x10<sup>6</sup> conidia /cm for biological control of the three land snails, *M. cantiana*, *T. pisana* and *S. putris*. results revealed that *B. bassiana* 3x10<sup>6</sup> ( Biofly ) was the highest toxic action against *M. cantiana* followed by *B. thuringiensis* var *Berliner* and *B. thorengensis* var *kurstaki* when using as a leaf dipping technique. Results of poison baits treatments of *B. thorengensis* var *Berliner* against various land snail species revealed that, *T. pisana* was the most sensitive to tested compounds (LC<sub>50</sub> 2.69 ppm) followed by *M. cantiana* (LC<sub>50</sub> 3.9 ppm), while *S. putris* snails were LC<sub>50</sub> 5.8.

In conclusion, control responsibility and / or the farmer must be taken into account, two important factors, where and when mollusciciding operations were carried out during the course of the present study. It is interesting to notice that terrestrial snails

were active only after irrigation, in moist weather, in damp evenings and early morning hours. It is thus under these circumstances that snails can come into contact with molluscicides. Also, the density of snails population are increases at field edges, especially beside field water canals, in tall herbs and under weeds which cover pontes, therefore, focal/ seasonal mollusciciding operations are likely to be the role rather than blanket (=area wide). Also, the present study indicated biological control plays an important role in reducing the population numbers of snail infestation minimize environmental pollution.

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