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EGYPTIAN ACADEMIC JOURNAL OF BIOLOGICAL SCIENCES TOXICOLOGY & PEST CONTROL



ISSN 2090-0791

WWW.EAJBS.EG.NET

Vol. 16 No. 2 (2024)

www.eajbs.eg.net



Efficacy and Joint Action of Some Biological and Safe Agents Against the Small Land Snail, *Helicella vestalis*, under Laboratory and Citrus Orchard Conditions

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REVIEWINFO

Review History Received:2/6/2024 Accepted:4/7/2024 Available:8/7/2024

Keywords: Biopesticides, land snails, control, clove oil, eugenol.

Six pesticides of different groups i.e. Acetamiprid 20% SP, Spinosad 24% SC, Carbendazim 50 % WP, Abamectin 2% + spirodiclofen18%, Copper sulphate 25% WP and Oxamyl 24 % SL) were evaluated against the small land snail, Helicella vestalis under laboratory and citrus field conditions. Experiments were conducted at the biological laboratory of Econ. Entomol. and Agric. Zoology Dept., Fac. of Agric., Menoufia Univ. and a citrus orchard at Al Bagour locality, Menoufia governorate, during spring 2023. Results indicated that the highest mortality was recorded with all tested agents when applied at the recommendation rates (100%), while it were 40, 30, 23, 33.3, 26.7, 33.3 % for Copper sulphate, Spinosad, Abamectin + Spirodiclofen, Carbendazim, Oxamyl, and Acetamiprid, respectively, while; clove oil 3% gave mortality of 23.3% and 53% when applied at a concentration of 5%, while, the highest mortality was recorded as 100% with the concentration of 10%. The highest mortality was recorded with the binary treatments of carbendazim + clove oil 5% and acetamiprid + clove oil 5%, when applied at half the recommended dose for each. While, copper sulphate + clove oil 5%, spinosad + clove oil 5%, abamectin + spirodiclofen + clove oil 5% recorded 96.67%. The highest snail mortality occurred 3 weeks after application with Acetamprid (86%) and clove oil 5% (81%), while the lowest rate was recorded with Oxamyl (69.4%). In field experiments, the highest mortality was recorded with the two binary treatments: acetamiprid + clove oil 5% (86.3%), followed by the oxamyl + clove oil 5% (83.3%), while it reached 83.3%, 77.6%, 79.1%, and 79.2% for abamectin + spirodiclofen + clove oil 5%, spinosad + clove oil 5% and carbendazim + clove oil 5%, respectively. Finally, the treatment of copper sulphate + clove oil 5% recorded the lowest one (68.1%) after three weeks of application. The chemical analyses of clove oil proved that it contains eugenol (62.7%) as the highest component and some other phenolic compounds that have been correlated to the many-sidedness of biological aspects. This search reported the prospective of clove oil and eugenol, as an efficient natural molluscicide replacement for its synthetic equivalent for snail control.

ABSTRACT

INTRODUCTION

Recently, land snails become an economic serious significant threat to sustainable agriculture in different parts of the world (Barker, 2002). Land snails have been classified as one of the most serious pests and public health, it damages most vegetables, field crops, fruit trees, orchards and ornamental plants (Ali *et al.*, 2020). Also, they lead to huge economic

damages because of their feeding (Hussein and Sabry, 2019). Also, they spoil agricultural products with their, faces and mucus, (Ali, 2017). The small land snail, *Helicella vestalis* is a threatening agricultural pest to orchard trees, such as Citrus causing damage to all tree parts (El-Okda *et al.*, 1989 a &b, El-Banhawy *et al.*, 1993). Therefore, the control of these land snails becoming very urgent, furthermore, until now chemical pesticides are still one of the most efficacious agents in land snail control at integrated pest management programs.

Currently, using chemical pesticides is not favourable. They cause disturbance of the biological control structure, unacceptable effects on other organisms, and build out resistance of pests to pesticides (Ismail *et al.*, 2015). In recent years, scientists have been focusing on studying the molluscicidal activity of various plants (Abdel-Rahman, 2017; Mortada *et al.*, 2012; Mourad, 2014) (Ismail *et al.*, 2015). This has led to increased interest in natural plant derivatives such as plant extracts and essential oils, which are known as botanical pesticides. These natural alternatives offer a way to reduce the use of chemical pesticides and can expand the range of environmentally friendly agents used in pest control, particularly essential oils (Edyta *et al.*, 2018). This study was conducted to shed light on the essential clove oil, *Syzygium aromaticum* separately and combined with the half dose of some chemical and bio-pesticides, if they give very close results in competing land snails to outline if clove oil can be used on plant hosts to reduce or control disease levels and stopping the development of land snails impact and minimizing the yield losses in quality and quantity.

MATERIALS AND METHODS

1-Tested Snails:

Healthy adults of the small land snail, *Helicella vestalis* were collected almost the same size and age from an infested untreated citrus orchard located at Al Bagour locality, Menoufia governorate, Egypt during spring 2023. The collected snails were transferred to the laboratory in plastic bags, then reared in a plastic box (40x30x30cm) covered with plastic containing certain holes filled with moist sandy loamy soil (10 snails/ box) and fed daily on green lettuce leaves were washed gently and dried placed on the box for 15 days to be acclimatized. under laboratory conditions of 25-27°C and 65-70% R.H. Snails with approximately the same age and shell diameter were starved for 24 h before treatments and then used in experiments. Dead snails were removed and only healthy snails were used in the following experiments.

2-Tested Pesticides:

Six pesticides of different chemical groups i.e. Acetamiprid 20% SP (Almared plus 200), Spinosad 24% SC (Tracer *Saccharopolyspora spinosa*), Carbendazim (Carbendazim 50 % WP), Abamectin 2% + spirodiclofen18% (Ignar 20% SC), Copper sulphate (Sulfo cup 25% WP) and Oxamyl (Vydate ® 24 % SL) were obtained from private companies for pesticides.

3-Essential Oil:

3.1.Sample Preparation for LC/MS-MS Analysis:

Clove essential oil was obtained from El-Gomhoria Company for oils and pharmaceutical Industries, in Cairo, Egypt. The oil was emulsified with 3% Tween 20. Oil emulsions were applied separately at concentrations of 3, 5 and 10 ml/L.

Oil emulsion (100 mg) was homogenized for 15 minutes with 800 ml of methyl alcohol 80%. After centrifugation for 10 minutes at 17,000 g, the supernatant was collected, and the extraction was done for the second time with methanol 80%.

The supernatants were collected and evaporated to the dryness.

The analysis of phenolic compounds by LC-MS/MS method was done with clove essential oil according to slightly modified methods.

3.2.LC-MS/MS Instrumentation and Analytical Conditions:

Analyses were carried out using an LCMS/MS 8050 system with a triple quadrupole mass spectrometer. The solution of samples was injected into a reversed-phase column (BEHC 8, 1.7 mm, 2.1 mm \times 150 mm, Waters, Milford, MA, USA) with appropriate precolumns under 40°C. The mobile phase consisted of a mixture of 10 mM formic acid (solvent A) and acetonitrile (solvent B) at a flow rate of 0.25 ml/minute. The linear gradient and isocratic flowers of the mobile phase were slightly modified as Gruz *et al.*, 2008 method. **4-Laboratory Tests:**

Treatments were conducted using the leaf spraying technique (Khalifa, 2015). Homogenous disks of lettuce leaves were sprayed in a series of the selected pesticides for 5 min and left for dryness. Two concentrations (recommended and half rates) of the tested compounds were prepared using diluted distilled water. Three ml of each concentration was spread on a Petri-dish surface with fresh green leaves of lettuce and moved in the circle for equal spreading leaving a residual layer of the tested compound. The treated lettuce disks were transferred into a plastic box and 10 adult snails were placed into. Each concentration had three replicates along with a control one. The dead snails were daily counted and removed daily. The corrected mortality was estimated by using Abbott's formula (Abbott, 1925).

5-Field Trials:

A citrus orchard in Al Bagour district, Menoufia governorate was utilized to perform field trials to assess the toxicity of the tested compounds against *H. vestalis*. An area of about one kirat (175 m^2) was chosen for the experiment. The orchard was irrigated 5 days before the treatment. The trial comprised of series of treatments including the control, in a randomized complete block design with four replicates (3 trees each) per treatment. A distance of 5 m was separated between each treatment.

The tested compounds were applied as a spray on the infested trees with a hand sprayer. The treatment began 5 days after irrigation and was performed every 7 days from the first application. The reduction of snail population before and after (7, 14 and 21 days) of treatment was the base for detecting the efficacy of each compound. Moreover, the mortality percentage was estimated using Henderson and Tilton (1955) formula.

Reduction%= 1- (treatment after /treatment before* control before/ control after)* 100 6- Statistical Analysis:

All data were computed by the ANOVA test using a computer program (Costat, 2008) to know Duncan's multiple range test and determine LSD 5% (least significant differences).

RESULTS

Laboratory Experiments:

The obtained results in Table (1) show the mean numbers of dead *Helicella vestalis* snails as affected by different agents for seven days.

The analysis of the obtained data in Table (1) recorded that there were significant variations in the numbers of dead individual snails seven days after treatment, at both recommended and half rates, among the tested agents compared to the control, furthermore, there were significant variations among the tested materials.

Regarding to data in Table (2) results reported that the highest mortality percentages were recorded with all tested agents applied at the recommendation rates (100%) in addition to clove oil 10 % also, gave a 100 % mortality percentage. As for the mortality percentages

of treated snails with half recommended rates of the applied agents, seven days of treatment, it were 40, 30, 23, 33.3, 26.7, 33.3 % for Copper sulphate 25% WP, Spinosad 24% SC, Abamectin 2% SC+ spirodiclofen18%, Carbendazim 50%WP, Oxamyl 24% SL, and Acetamiprid 20% SP, respectively. While clove oil 3 % gave a 23.3 % mortality percentage.

Table 1: Effect of recommended and half rates of the tested agents on the small land snail,

 Helicella vestalis under laboratory conditions

					C 1	1 77 7.	11		
Tested agents & Concentrations		The mean numbers of dead <i>Helicella vestalis</i>							
		snails/30 individuals							
		Days after treatment							
	-	1 st	2 nd	3 rd	4 th	5 th	6 th	7 th	
Copper sulphate 25%	Recom.	1	9	14	21	28	30	30 a	
WP	Half	0	2	5	7	11	12	12 c	
Spinosed 24% SC	Recom.	1	10	15	20	28	29	30 a	
Spinosad 24% SC	Half	0	1	3	7	8	8	9 de	
Abamectin 2% SC+	Recom.	1	7	17	23	28	30	30 a	
spirodiclofen18%	Half	0	2	2	5	6	7	7 e	
Carbendazim 50%WP	Recom.	1	6	14	19	24	28	30 a	
Carbendazini 50% wr	Half	0	2	3	5	7	9	10 d	
Oxamyl	Recom.	0	4	16	20	26	29	30 a	
24% SL	Half	0	1	3	3	4	7	8 de	
Acetamiprid	Recom.	4	25	29	30	30	30	30 a	
20% SP	Half	0	0	1	4	7	8	10 d	
	3%	0	0	1	4	4	7	7 e	
Clove oil	5%	1	5	9	10	11	13	16 b	
	10%	2	20	27	29	30	30	30 a	
Control		0	0	0	0	0	0	0 f	
LSD 5%		-	-	-	-	-	-	1.99	

means in column followed by different letter(s) are significantly differences at 5% level

Table 2: Efficacy of two rates of tested agents on the small land snail, *H. vestalis* under laboratory conditions

		Mortality percentages of H. vestalis snail							
Tested agents	Days after treatment								
& Concentrations	1 st	2 nd	3 rd	4 th	5 th	6 th	7 th		
Copper sulphate25%	Recom.	3.3	30.0	46.7	70.0	93.3	100.0	100.0	
WP	Half	0.0	6.7	16.7	23.3	36.7	40.0	40.0	
Spinosad 24% SC	Recom.	3.3	33.3	50.0	66.7	93.3	96.7	100.0	
Spillosad 24% SC	Half	0.0	3.3	10.0	23.3	26.7	26.7	30.0	
Abamectin 2% SC	Recom.	3.3	23.3	56.7	76.7	93.3	100.0	100.0	
+spirodiclofen18%	Half	0.0	6.7	6.7	16.7	20.0	23.3	23.3	
Carbendazim	Recom.	3.3	20.0	46.7	63.3	80.0	93.3	100.0	
50% WP	Half	0.0	6.7	10.0	16.7	23.3	30.0	33.3	
Oxamyl 24% SL	Recom.	0.0	13.3	53.3	66.7	86.7	96.7	100.0	
Oxalliyi 24% SL	Half	0.0	3.3	10.0	10.0	13.3	23.3	26.7	
Acetamiprid 20% SP	Recom.	13.3	83.3	96.7	100.0	100.0	100.0	100.0	
Acetainipilu 20% Sr	Half	0.0	0.0	3.3	13.3	23.3	26.7	33.3	
	3%	0.0	0.0	3.3	13.3	13.3	23.3	23.3	
Clove oil	5%	3.3	16.7	30.0	33.3	36.7	43.3	53.3	
	10%	6.7	66.7	90.0	96.7	100.0	100.0	100.0	

The obtained results in Table (3) show the mean numbers of dead *Helicella vestalis* snails as affected by the combined action of the different agents for seven days.

Analysis of the obtained data in Table (3) recorded that there were significant differences in the numbers of dead individuals of snails, seven days after treatment, at the binary of the half rates, between the tested agents except oxamyl + clove oil 5%.

Regarding to the results in Table (4) revealed that the highest mortality (100%), after seven days, was recorded with the treatments of Carbendazim 50% WP+ Clove oil 5%, Acetamiprid 20% SP + Clove oil 5%, when applied binary at the half recommended compared to control treatment. While, the treatments of Copper sulphate25% WP + Clove oil 5%, Spinosad 24% SC + Clove oil 5%, and Abamectin 2% SC+ spirodiclofen18%+ Clove oil 5%, similarly, recorded 96.67%, seven days after treatment, and the lest mortality percentage was registered with the binary of Oxamyl 24%SL+ Clove oil 5% as 86.67%, seven days after treatment.

		Mean number of dead snails of <i>H. vestalis</i> /30 individuals						
Tested agents & Concentrations	Days after treatment							
	1 st	2 nd	3 rd	4 th	5 th	6 th	7 th	
Copper sulphate 25% WP + Clove oil 5%	2	4	11	17	24	28	29 a	
Spinosad 24% SC + Clove oil 5%	2	4	11	17	23	28	29 a	
Abamectin 2% SC+ spirodiclofen18%+ Clove oil 5%	1	4	7	14	24	26	29 a	
Carbendazim 50% WP+ Clove oil 5%	2	4	11	16	22	27	30 a	
Oxamyl 24%SL+ Clove oil 5%	1	2	4	8	17	23	26 b	
Acetamiprid 20% SP + Clove oil 5%	3	7	13	23	29	30	30 a	
Control	0	0	0	0	0	0	0 c	
LSD 5%	-	-	-	-	-	-	2.73	

Table 3: Joint action of the tested agents (half rates) on the small land snail, *Helicella* vestalis under laboratory conditions.

means in column followed by different letter(s) are significantly differences at 5% level

Table 4: The combined effect of some safe agents on the small land snail, *Helicella vestalis*, under laboratory conditions

Tested agents & Concentrations		Mortality % of <i>H. vestalis</i> snail							
		Days after treatment							
	1 st	2 nd	3 rd	4^{th}	5^{th}	6 th	7^{th}		
Copper sulphate25% WP + Clove oil 5%	6.67	13.33	36.67	56.67	80.00	93.33	96.67		
Spinosad 24% SC + Clove oil 5%	6.67	13.33	33.33	56.67	76.67	93.33	96.67		
Abamectin 2% SC+ spirodiclofen18%+ Clove oil 5%	3.33	13.33	23.33	46.67	70.00	86.67	96.67		
Carbendazim 50% WP+ Clove oil 5%	6.67	13.33	36.67	53.33	73.33	90.00	100.00		
Oxamyl 24%SL+ Clove oil 5%	0.00	6.67	13.33	26.67	56.67	76.67	86.67		
Acetamiprid 20% SP + Clove oil 5%	10.00	23.33	43.33	76.67	96.67	100.00	100.00		

Field Experiments:

The obtained results in Table (5), show the efficacy of some biopesticides and chemical agents against the small land snail, *Helicella vestalis* infected citrus trees, under open field conditions as well as reduction %.

As for the statistical analysis of the mean numbers of live snails as affected by the treatment of infected citrus trees with different pesticides over three weeks Table (5) there were significant variations in the numbers of live individuals of snails, three weeks after treatment between all tested materials and control treatment. As well as there were

significant differences in the numbers of live individuals of the snail between the treatments of Acetamiprid 20% SP & Clove oil 5% and other tested materials.

Regarding to the obtained results in Table (5), the highest mortality %, after three weeks of application was recorded with the treatments of Acetamiprid 20% SP (86%) and Clove oil 5% (81%), while the treatments of Abamectin 2% SC+ spirodiclofen18% & Carbendazim 50% WP & Spinosad 24% SC recorded mortality percentages as (76.4%) (75.5%) (74.5%) respectively. The lowest mortality percentage was recorded with the treatment of Oxamyl 24% SL as (69.4%).

	Mean no. of live snails of <i>H. vestalis</i> / three trees (Reduction %)					
Tested agents & rates	Periods after treatment					
	Pre- treatment	1 week	2 weeks	3 weeks		
Copper sulphate 25% WP	236	198 (19.5)	118 (50.1)	86 b (67.7)		
Spinosad 24% SC	259	208 (12.3)	141 (45.7)	67 cd (74.5)		
Abamectin 2% SC+ spirodiclofen18%	263	212 (22.6)	163 (38.2)	63 cd (76.4)		
Carbendazim 50% WP	270	216 (23.2)	110 (59.4)	67 cd (75.5)		
Oxamyl 24% SL	242	208 (17.5)	143 (41.1)	75 bc (69.4)		
Clove oil 5%	265	209 (24.3)	104 (60.9)	51 de (81.0)		
Acetamiprid 20% SP	263	180 (34.3)	98 (62.8)	40 e (85.0)		
Control	259	270	260	263 a		
LSD 5%	-	-	-	18.0		

Table 5: Efficacy of some safe and bioagents against the small land snail, *Helicella vestalis* under open field conditions as well as reduction %

means in column followed by different letter(s) are significantly differences at 5% level

The obtained results in Table (6), show the combined effect of some biopesticides and chemical agents against the small land snail, Helicella vestalis infected citrus trees, under open field conditions as well as reduction % along three weeks of application.

Results in Table (6), were statistically analysed and reported that there were significant variations in the numbers of live individuals of snails, three weeks after treatment between all tested materials and control treatment. Furthermore, there were significant differences in the numbers of live individuals of snails, three weeks after treatment between the treatments of Copper sulphate 25% WP + Clove oil 5% and Acetamiprid 20% SP + Clove oil 5% treatments. While, there were no significant differences in the numbers of live individuals of snails, three weeks after treatment, among Oxamyl 24% SL+ Clove oil 5% (49 individuals) & Carbendazim 50% WP+ Clove oil 5% (58 individuals) & Spinosad 24% SC + Clove oil 5% (63 individual) & Abamectin 2% SC+ spirodiclofen 18% + Clove oil 5% (60 individual) treatments (LSD 5% = 14.2).

With regard to results in Table (6) the highest mortality %, after three weeks of application was recorded with the treatments of Acetamiprid 20% SP + Clove oil 5% (86.3%), followed by the binary treatments of Oxamyl 24%SL+ Clove oil 5% (83.3%), while it was (77.6%), (79.1%) and (79.2%) for Abamectin 2% SC+ spirodiclofen18%+ Clove oil 5%, Spinosad 24% SC + Clove oil 5% and Carbendazim 50% WP+ Clove oil 5%, respectively. Finally, the binary treatment of Copper sulphate 25% WP + Clove oil 5% recorded the lowest reduction percentage (68.1%) after three weeks of application.

Hencena vestans under open field conditions									
	I	Mean number	s of live snai	ls &					
	of <i>H. vestalis</i> nail								
		(Reduction %)							
Tested agents & rates		Periods at	fter treatment	t					
	Pre- treatment	1 week	2 weeks	3 weeks					
Copper sulphate25% WP + Clove oil 5%	263	231 (18.9)	160 (45.9)	98 b (68.1)					
Spinosad 24% SC + Clove oil 5%	258	220 (21.3)	128 (55.9)	63 c (79.1)					
Abamectin 2% SC+ spirodiclofen18%+ Clove oil 5%	230	224 (10.1)	144 (44.3)	60 c (77.6)					
Carbendazim 50% WP+ Clove oil 5%	244	229 (13.3)	163 (40.6)	58 c (79.2)					
Oxamyl 24% SL+ Clove oil 5%	252	239 (12.5)	134 (52.7)	49 cd (83.3)					
Acetamiprid 20% SP + Clove oil 5%	244	205 (22.5)	120 (56.3)	39 d (86.3)					
Control	240	260	270	280 a					
LSD 5%	-	-	-	14.2					

Table 6: The combined effect of different safe and bioagents on the small land snail, Helicella vestalis under open field conditions

means in column followed by different letter(s) are significantly differences at 5% level

Results shown in Table (7), indicate that clove oil has high amounts of Eugenol (62.72 mg/ml), Eugenol acetate (19.1 mg/ml), it also has B-Caryophyllene (11.5 mg/ml), Alpha-Humulene (1.77 mg/ml), Alpha-(E)-Ionone (2.8 mg/ml) and Alpha-Cubebene (0.53 mg/ml), it could be concluded that the Phenolic compounds of the tested oil especially Eugenol and Eugenol acetate play important role in the process of snail control. The obtained results are in harmony with those of Bauer *et al.* (2001), Soha *et al.* (2015), and Abdel-Rahman (2017, 2020).

Phenolic compounds	Content (mg/100 ml oil sample)
Eugenol	62.72
Alpha- Copaene	1.14
Eugenol acetate	19.10
B-Caryophyllene	11.50
Delta-Cadinene	0.21
Alpha-Humulene	1.77
Alpha-(E)-Ionone	2.8
Alpha-Cubebene	0.53

Table 7: Chemical analysis of phenolic compounds in the clove oil.

DISCUSSION

The obtained results are consistent with those of Genena and Mostafa (2008) who conducted tests on six pesticides i.e. bensultap, chlorpyrifos-ethyl, deltamethrin, diazonixy, lambda-cyhalothrin, and methomyl in the laboratory for their effects against *M. cantiana* and *E. vermiculata*, using poison baits at an incessant concentration of 2%, where the results reported that all pesticide baits had a toxic effect, with increasing of the mortality percentages with the increasing the period of exposure, moreover, deltamethrin registered the highest toxicity affect 70.0% and 93.3% on *M. cantiana* and *E. vermiculata*, respectively, after three days, meanwhile, methomyl, recorded 100.0% mortality after 7 and 12 days of

M. cantiana and E. vermiculata, respectively, and finally, chlorpyrifos-ethyl occupied the lowest mortality rank against the two land snails.Ismail and Abd El-Kader (2011) conducted an evaluation on flower-bud powder and eugenol from Syzygium aromaticum in controlling juveniles and adults of *Monacha cartusiana* using a baiting technique. They tested three concentrations (10%, 20%, and 40%) of bud-flower and (1%, 2%, and 4%) of eugenol under laboratory and field conditions. At the lowest concentrations of 10% for clove-bud powder and 1% for eugenol, mortalities were 67.5%, 70%, 75%, and 47.5% respectively, however, higher concentrations of 40% for clove-bud powder and 4% for eugenol, gave 99%, 100%, 100%, and 97.5% mortalities for juvenile and adult snails 21 days after treatment under laboratory conditions, while in field conditions, the mortalities were 39.6%, 57.2%, and 62.4% for adult snails at the three tested concentrations of clove-bud powder, respectively. Hegab et al. (2013) evaluated methomyl and copper sulphate on Eobania vermiculata and Helicella vestalis snails under both laboratory and field conditions, where the laboratory tests reported that the mortality percentages of the snails were raised with higher concentrations and longer exposure durations, in addition, as for *Eobania vermiculata*, all concentrations of methomyl resulted 85%, 92.5%, and 100% with concentrations of 1%, 3%, 5%, and 7% mortalities, after 7 days post-treatment, meanwhile, copper sulphate recorded 65%, 70%, 80%, and 85%, respectively, while methomyl was more effective against H. vestalis snails, resulting in 100-100% mortality, compared to copper sulphate, which resulted in 47.6-72.5% mortality 7 days post-treatment, while under field experiment, the reduction percentages were very low, as 7.55% and 1.13% for methomyl and copper sulphate against E. vermiculata snails. Soha et al. (2015) studied the effect of clove (Syzygium aromaticum) extract against land slug, Limax flavus, under laboratory and field conditions, where slugs were treated with different concentrations of plant extract as a contact for one week and LC₅₀ value was calculated and revealed that plant extract by ethanol was more toxic than that by acetone. Samah M. Abdel-Kader et al. (2016) tested metaldehyde 5%, methomyl 20% SL, methomyl 24% SL, methiocarb 2% RB(G), and Bacillus thuringiensis 6.5% WP against the land snail, Monacha cartusiana under field conditions and found that metaldehyde, methomyl 20%, methomyl 24%, and methiocarb baits were the most effective in reducing snail numbers over the 21-day experimental period, while Bacillus thuringiensis was less effective. Wafaa A. Shahawy (2018) studied the biochemical effects of two molluscicides: Agrinate and Biomagic on two land snail species: Helicella vestalis and Theba pisana, as well as the negative effects of sub-lethal concentration (1/4 LC₅₀) of both molluscicides on aspartate aminotransferase, alanine aminotransferase, total lipids and total proteins. Abdel-Rahman (2020) tested the molluscicidal action of clove oil, Syzygium aromaticum, black cumin oil or black seed oil, Nigella sativa, and mustard oil, Brassica alba along with neomyl and reported that tested compounds were effective against the Monacha snail, and neomyl exhibited the highest molluscicidal effect, followed by S. aromaticum oil, N. sativa oil, and B. alba oil under laboratory conditions, in addition, mortality percentages after 21 days of treatment was 90%, 79%, 66%, and 60% for neomyl, S. aromaticum, N. sativa, and B. alba, respectively. Mona Abou El Atta (2023) investigated the molluscicidal activity of the Ali and Doaa monoterpene, agrinate, thymol and thyme oil against adult land snails, Succinea putris and Eobania vermiculata, and found that Agrinate was the most effective, followed by thymol, while thyme essential oil had the lowest effect on the adult S. putris and E. vermiculata, additionally, thymol was found to prevent mold formation and exhibit antifungal activity in poisonous bait. Radwan and El-zemity (2007) reported that eugenol, found in S. aromaticum, was highly toxic to *L. acuminata* (LC₅₀ 96h = 1.41 mg/L). Moreover, eugenol showed high toxicity to the schistosomiasis-carrying aquatic snail, Biomphalaria alexandrina, with an LC₅₀ of 28 mg/L. Recently, Amira et al. (2023) reported significant sub-lethal effects of clove oil or eugenol at 48 and 72 hours, including a notable increase in reduced glutathione (GSH) levels. Hend Sh. Ghareeb (2024) assessed the molluscicidal effects of crude juice of Citrus limon fruit and Citrus limon peel on Eobania vermiculata and Monacha cartusiana stages, and found that the juveniles of three months old were more susceptible to C. limon fruit and C. limon peel juices than the other ages of both snails, while in the field the reduction of both snail adult individuals were 70.39 & 62.30% and 62.08 & 55.79%, consecutively. Finally, El-Sayed et al. (2024) evaluated the efficiency of methomyl and metaldehyde as well as a biocide Biogard® (Bt), at recommended field rates against the land snail, E. vermiculata under field conditions in guava orchard at Alexandria Governorate, and found that all tested compounds exerted significant reduction in the number of living snails on guava trees one day after treatment and until the end of the experiment compared to untreated trees, where the three tested compounds were found to be toxic to E. vermiculata as 79.39, 69.12 and 86.57% for methomyl, Bt and metaldehyde after 21 days of experiment, respectively, moreover the average initial kill were 28.67, 16.39 and 41.66% and average residual effect were 67.98, 59.55 and 78.81 % for methomyl, Bt and metaldehyde, respectively.

Finally, it could be concluded that all tested materials gave adequate control of the target snail when applied singly or binary, especially the safe materials such as Clove oil 5% and Copper sulphate 25% WP.

Declarations:

Ethical Approval: Not applicable

Competing interests: The author declares that no duality of interest associated with this manuscript.

Funding: No specific funding was received for this work

Availability of Data and Materials: All datasets analysed and described during the present study are available from the corresponding author upon reasonable request.

Acknowledgements: Not applicable.

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ARABIC SUMMARY

الكفاءة والتأثير المشترك لبعض المواد البيولوجية والآمنه ضد القوقع الصغير هيلاسيلا فيستالس تحت ظروف المعمل وبستان الموالح

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محموعات كيميائية مختلفة هي (Acetamprid 20% SP، تم تقييم فاعلية سنة مبيدات تنتمي إلى مجموعات كيميائية مختلفة هي (Abamectin 2% + spirodiclofen 18% ،Carbendazim 50% WP ،Spinosad 24% SC ،Copper sulphate 25% WP تحت Helicella. vestalis صد القوقع الصغير المعملية والحقلية للموالح .

أجريت التجارب في المعمل البيولوجي التابع لقسم الحيوان الزراعي ، كلية الزراعة جامعة المنوفية وبستان موالح بمركز الباجور بمحافظة المنوفية خلال ربيع عام 2023.

وأشارت النتائج إلى تسجيل أعلى نسبة موت لجميع المواد عند تطبيقها بالنسب الموصى بها (100%) بينما بلغت 40، 30، 23، 33.3، 26.7، 33.3% لكبريتات النحاس، سبينوساد ، أبامكتين + سبيروديكلوفين ، كاربندازيم ، أوكساميل ، وأسيتاميبريد ، على التوالي ، بينما أعطى زيت القرنفل 3% معدل وفيات 23.3% ، 53% عند تطبيقه بتركيز 5% ، في حين سجلت أعلى نسبة موت 100% عند تركيز 10%. تم تسجيل أعلى معدل موت مع معاملات الكاربندازيم + زيت القرنفل 5% والأسيتاميبريد + زيت القرنفل 5% ، عند تطبيقها بنصف الجرعة الموصى بها لكل منهما. بينما سجلت كبريتات النحاس + زيت القرنفل 5% ، سبينوساد + زيت القرنفل 5% ، أبامكتين + سبيروديكلوفين + زيت القرنفل 5% والأسيتاميبريد + زيت القرنفل 5% ، عند تطبيقها بنصف الجرعة الموصى بها لكل منهما. بينما سجلت كبريتات النحاس + زيت القرنفل 5% ، سبينوساد + زيت بنصف الجرعة الموصى بها لكل منهما. بينما سجلت كبريتات النحاس با زيت القرنفل 5% ، معدل لموت القواقع بنصف الجرعة الموصى بها لكل منهما. وينما سجلت كبريتات النحاس بازيت القرنفل 5% ، معدل موت القواقع القرنفل 5% ، أبامكتين + سبيروديكلوفين بازيت القرنفل 5% نسبة 66.6%. سجل أعلى معدل لموت القواقع بعد 3 أسابيع من تطبيق الأسيتامبريد (86%) وزيت القرنفل 5% القرن ، في حين تم تسجيل أدنى معدل مع الأوكساميل (69.4%).

وفي التجارب الحقلية سجلت أعلى نسبة موت مع المعاملات : الأسيتامبريد + زيت القرنفل 5٪ (86.3٪) ، يليهما الأوكساميل + زيت القرنفل 5٪ (83.3٪) ، فيما بلغت 83.3٪ ، 77.6٪ ، 79.1. 79.2٪ لمعاملات أبامكتين + سبيروديكلوفين + زيت القرنفل 5٪، سبينوساد + زيت القرنفل 5٪، وكربندازيم + زيت القرنفل 5٪ ، على التوالي. وأخيراً سجلت معاملة كبريتات النحاس + زيت القرنفل 5٪ أدنى نسبة (68.1٪) بعد ثلاثة أسابيع من التطبيق.

أثبتت التحاليل الكيميائية لزيت القرنفل أنه يحتوي على الأوجينول (62.7٪) كأعلى مكون وبعض المركبات الفينولية الأخرى التي ارتبطت بمجموعة متنوعة من الأنشطة البيولوجية. يسلط هذا البحث الضوء على إمكانات زيت القرنفل والأوجينول، كبديل طبيعي فعال كمبيد الرخويات لنظير اته الكيميائية لمكافحة القواقع