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ORIGINAL ARTICLE

Potential molluscicidal activity of the aqueous extracts of some plants and their powders against terrestrial snail *Monacha obstructa* (L. Pfeiffer, 1842) under laboratory and field conditions

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Abstract

As it is known that the excessive use of pesticides causes many environmental problems, the effects of four aqueous plant extracts and their powders [*Solanum nigrum* L., *Withania somnifera* (L.) Dunal, *Salix mucronata* Thunb. and *Lawsonia inermis* L.] were evaluated as natural molluscicides on the land snail *Monacha obstructa* (Family: Hygromiidae) under laboratory conditions. Three different bioassay methods were used: contact, leaf-dipping and bait techniques. The results indicated that, using all methods, *S. Nigrum* extract was the most toxic extract for the terrestrial snail *M. obstructa*. The contact technique of the tested plant extracts was the most effective method of application compared to other methods. Moreover, using plant extracts was better than using powders of these plants for controlling the terrestrial snail *M. obstructa*. In addition, the results indicated that the aqueous extract of *S. nigrum* gave the highest percentage of reduction in the snail population when assessed under field conditions. The obtained data showed that plant extracts were significantly effective against the terrestrial snail and could be used as alternatives to pesticides in integrated pest management.

Keywords: *Monacha obstructa*, pest management, plant extracts, *Solanum nigrum*, the clover snail

Introduction

Land snails are destructive agricultural pests causing economic damage to a wide variety of plants, including vegetables, forage crops, tree fruits, shrubs, flowers, green ground cover and newly sown lawn grasses. Moreover, they play an important role in transmitting and spreading diseases to cultivated plants (Godan 1983; Baker 1989). The clover land snail, *Monacha obstructa,* is the most common and serious pest in Egypt, causing substantial damage to different agricultural crops in various governorates. Recent studies have indicated that this pest was the most abundant snail in some Upper Egypt governorates, in Assiut governorate (Ibrahim *et al.* 2020; Abo-Elnaser 2022) and in Sohag governorate (Mahmoud *et al.* 2021). The plant kingdom is a rich source of potential pest control. Active substances extracted from plants are considered to be natural alternatives and are being researched to find new, safe and effective chemicals which act as toxicants. The toxic effect of plant extracts against the land snail *Monacha* sp. has been reported by many researchers) Farag 2017; Ibrahim *et al.* 2022). El-Sherbini *et al.* (2009) studied the molluscicidal activity of *Solanum nigrum* against *Biomphalaria alexandrina* (Ehrenberg). They found that extracts from mature leaves of *Solanum* species exhibited molluscicidal activities. The highest recorded mortality rate was observed in the *S. nigrum* extract, followed by *S. sinaicum* Boiss., while *S. villosum* Mill. had the lowest mortality rate. Also, environmental problems and toxic effects on non-target organisms and beneficial invertebrates resulting

from extensive pesticides use have prompted scientists to search for alternative methods of snail management, including safe molluscicides with different modes of action. The objective of the present investigation was to study the molluscicidal effect of plant extracts against terrestrial snail *M. obstructa*, as a new approach in integrated pest management programs.

Materials and Methods

Plants used

The plants used in this study were collected from different regions of Sohag governorate, Egypt. The scientific name, family, English name and part used are shown in the following Table 1.

Preparation of powders and extracts of plants

For the tests the plant part was dried at room temperature (26 ± 3°C) and then ground into fine powder using an electric grinder. Aqueous extracts of plants were prepared according to the method of Souza *et al.* (2013). The concentrations w/v % (the percent of weight of solute in the total volume of solution) of each technique were obtained by mixing dry powder with distilled water. The powders were soaked in distilled water for 72 hours. The suspended solutions were then filtrated and autoclaved with steam under pressure at 121°C for 20 minutes. The resulting aqueous solutions were used for the tests.

Laboratory evaluation

Tested snails

Adult *M. obstructa* land snails, with a shell diameter of about 12 mm, were obtained from different infested nurseries in Sohag governorate. The obtained snails were transferred in plastic bags to the laboratory of the Agriculture Zoology and Nematology Department, Faculty of Agriculture, Al-Azhar University, Assiut Branch, then transferred to plastic containers containing moist sterilized sandy loam soil 1:1 (v:v) and fed fresh lettuce leaves (*Lactuca sativa* L.) for 14 days to be laboratory acclimatized.

Application methods

Aqueous extracts from the tested plants were examined using contact and leaf-dipping techniques, while the dried powder of these plant parts was assessed using the poisonous bait technique to investigate their toxicity against the land snail *M. obstructa.*

Contact technique

The residual film technique was conducted to study the effect of four concentrations of aqueous plant extracts: 2.5, 5, 10 and 20% (w/v). Two ml of each concentration were deposited and distributed on the bottom of a Petri dish by gently moving the dish in circles. The water evaporated under room conditions (23°C ± 2) in a few minutes leaving a thin layer of film of the different concentrations for each plant extract (Abdel-Kader 2020). Three Petri dishes (9 cm in diameter) were used for each concentration. Ten adult snails with the same shell diameter (12 mm ± 1) were introduced into each Petri dish. In addition, a parallel control test was conducted using plain water. Observation of mortality was conducted using a stainless steel needle according to El-Okda (1981).

Leaf dipping technique

Similar pieces of green lettuce leaves (25 cm^2) were dipped in glass jars containing 100 ml of each plant extract for 5 seconds at four concentrations: 2.5, 5, 10 and 20 % (w/v) for each plant extract. The leaves were air-dried (23°C ± 2) and then introduced to land snails in three plastic boxes (3/4 kg capacity) for each concentration. Each box contained 1/2 kg of moisturized and sterilized clay. Additionally, a parallel control test was conducted using plain water. Ten adult snails with the same shell diameter (12 mm ± 1) were introduced into the boxes, which were then covered with muslin cloth held by rubber bands.

Poisonous bait technique

Four concentrations, 2.5, 5, 10 and 20% (w/w), were used as poisonous baits. These baits were prepared by incorporating the appropriate weight of the tested material with wheat bran and five parts of black sugarcane syrup to give 100 parts of poisonous bait. The bait was moistened with an appropriate amount of water to form a crumbly mash mixture (Mortada *et al.* 2012). Twenty grams of each bait were spread on the

Table 1. Scientific name, family, English name and part of plant used for the extracts

Scientific name	Family	English name	Used part
Solanum nigrum L.	Solanaceae	black nightshade	fruits
Lawsonia inermis L.	Lythraceae	Egyptian privet	eaves
Withania somnifera (L.) Dunal	Solanaceae	ashwagandha	fruits and leaves
Salix mucronata Thunb.	Salicaceae	safsaf willow	leaves

bottom of each plastic box (3/4 kg capacity) containing optimal soil. Three replicates of 10 adults for each concentration were used, in addition to the untreated check. Each box was then covered with a muslin cloth held by rubber bands.

Data analysis

Mortality percentages were calculated 1, 7, 14 and 21 days post treatment and were corrected using Abbott's formula (1925), then subjected to statistical analysis using SPSS statistics 22 to determine the lethal concentration causing 50% mortality (LC_{50}). Furthermore, we used Costat Statically computer program to determine LSD test at a significance level of 0.05.

Field evaluation

Among the four plant extracts tested in the laboratory, the two most toxic extracts, *S. nigrum* and *W. somnifera*, were chosen to be evaluated at a concentration of 20% (w/v) for the spray technique and 30% (w/w) for the bait technique against *M. obstructa* under field conditions. The trial was performed in Arab Sabha village, Dar El-Salam district, Sohag governorate, in April 2023. The spraying method was used in an experimental area of two feddans (about 8400 m^2) cultivated with orange and mandarin trees and was heavily infested with *M. obstructa* snails. The orchard was irrigated 4 days before treatment. Five trees were randomly chosen for each treatment, and three replicates were used per treatment. Three treatments were conducted including the control (Mostafa 2020).

Using the bait method, the experiment was conducted in an Egyptian clover field heavily infested with *M. obstructa* snails in Naqnaq village, Dar El-Salam district, Sohag governorate. An experimental area of two feddans (about 8400 m^2) was divided into three plots including the control. Each plot was divided into three subplots (three replicates). Poison baits were prepared as follows: the amount of tested material + appropriate weight from bran + 5% sugar cane syrup (as attractant substance) to give 100 parts of bait, about 200 grams of prepared poisonous baits were offered on plastic sheets 50 × 50 cm (El-Sayed 2010), and distributed in the experimental plots at known distances (in the middle and corners of each subplot). Live snails per 0.25 m^2 were recorded in check and treatments before application and 1, 7, 14 and 21 days post-treatment. Population reduction percentages were calculated according to the formula given by Henderson and Tillton (1955):

$$\% \text{ Reduction} = \frac{1 - (t2 \times r1)}{(t1 \times r2)} \times 100,$$

where r1 – number of live snails before treatment in untreated plots; r2 – number of live snails after treatment in untreated plots; t1 – number of live snails before treatment in treated plots; t2 – number of live snails after treatment in treated plots.

Results

Laboratory evaluations

Efficiency of some aqueous plant extracts on *Monacha obstructa* snails using the residual film technique

Data in Table 2 show the mortality percentages of the land snail, *M. obstructa* after different periods of exposure to aqueous plant extracts using the residual film technique. Results indicated that, *Solanum nigrum* extract was the most toxic, with mortality percentages 60.00 and 86.66% after 21 days at 10 and 20% concentrations, respectively, followed by *Withania somnifera*, *Salix mucronata* and *Lawsonia inermis*, with mortality percentages of 53.34 and 70.00%; 50.00 and 60.00% and 33.34 and 43.34% at 10 and 20% concentrations, respectively after 21 days. In general, it was clear that the tested aqueous extracts reduced *M. obstructa* population density to low values after 7 days of treatment. Thereafter, the mortality percentages increased gradually and were maximum after 21 days.

Efficiency of aqueous plant extracts on *Monacha obstructa* snails using the leaf dipping technique

The mortality percentages of some aqueous plant extracts against *M. obstructa* using the leaf dipping technique after 1, 7, 14 and 21 days are presented in Table 3. The results showed that the mortalities increased with increasing plant extract concentrations and exposure time. The highest mortality percentages of *M. obstructa* were recorded with *S. nigrum* extract, followed by *W. somnifera* extract, with values of 66.67 and 56.67% at 20% concentration 21 days post-treatment, respectively. While the extracts of *S. mucronata* and *L. inermis* had the least effect, with mortality percentages of 50.00 and 36.67% at the same previous concentration 21 days post-treatment, respectively. The lowest mortality percentages were detected 1 day post-treatment then increased gradually.

Efficiency of some plant powders on *Monacha obstructa* using the poisonous bait technique

The effect of four plant powders against *M. obstructa* using the bait technique after 1, 7, 14 and 21 days of treatment was recorded in Table 4. The results showed

Table 2. Mortality percentages of aqueous plant extracts against *Monacha obstructa* using the residual film technique

Plant extracts	Con. [%]	Mortality percentages [days]				General mean
		1	7	14	21	
Solanum nigrum L.	2.5	13.34	30.00	33.34	40.00	29.17
	5	26.67	36.67	40.00	43.34	36.67
	10	26.67	46.67	46.67	60.00	45.00
	20	33.34	60.00	86.67	86.67	66.67
Withania somnifera (L.) Dunal	2.5	10.00	20.00	23.34	33.34	21.67
	5	13.34	26.67	33.34	36.67	27.50
	10	16.67	36.67	43.34	53.34	37.50
	20	23.34	53.34	56.67	70.00	50.84
Salix mucronata Thunb.	2.5	0.00	6.67	10.00	13.34	7.50
	5	3.34	10.00	16.67	16.67	11.67
	10	10.00	30.00	30.00	50.00	30.00
	20	16.67	50.00	53.34	60.00	45.00
Lawsonia inermis L.	2.5	0.00	0.00	3.34	3.34	1.67
	5	0.00	13.34	23.34	23.34	15.00
	10	6.67	16.67	30.00	33.34	21.67
	20	10.00	30.00	33.34	43.34	29.16
LSD	(0.05)	5.95	11.73	14.09	15.18	12.73

Table 3. Mortality percentages of aqueous plant extracts against *Monacha obstructa* using leaf dipping

Plant extracts	Con. [%]	Mortality percentages [days]				General mean
		1	7	14	21	
Solanum nigrum L.	2.5	0.00	3.34	3.34	13.34	5.00
	5	0.00	10.00	10.00	20.00	10.00
	10	3.34	13.34	13.34	26.67	14.18
	20	6.67	33.34	40.00	66.67	36.67
Withania somnifera (L.) Dunal	2.5	0.00	6.67	6.67	10.00	6.67
	5	3.34	6.67	10.00	16.67	8.33
	10	6.67	16.67	16.67	26.67	16.67
	20	10.00	33.34	46.67	56.67	36.67
Salix mucronata Thunb.	2.5	0.00	0.00	0.00	3.34	0.84
	5	0.00	6.67	13.34	16.67	9.17
	10	0.00	6.67	16.67	20.00	10.83
	20	6.67	20.00	33.34	50.00	27.50
Lawsonia inermis L.	2.5	0.00	0.00	0.00	0.00	0.00
	5	0.00	3.34	3.34	6.67	3.34
	10	0.00	10.00	13.34	13.34	9.17
	20	6.67	20.00	23.34	36.67	21.67
LSD	(0.05)	3.63	8.70	11.60	15.44	10.50

that mortalities increased with increasing plant powder concentration and exposure time. The highest mortality percentages of *M. obstructa* were recorded with *S. nigrum* baits, followed by *W. somnifera* with values 36.67; 53.34 % and 23.34; 43.34 % at 10 and 20% concentrations 21 days post-treatment, respectively. The effects of *S. mucronata* and *L. inermis* baits were the least with mortality percentages of 20.00; 36.67% and 13.34; 33.34% at the same previous concentrations 21 days post-treatment, respectively. The lowest

Table 4. Mortality percentages of some plant powders against *Monacha obstructa* using the poisonous bait technique

Plant extracts	Con. [%]	Mortality percentages [days]				General mean
		1	7	14	21	
Solanum nigrum L.	2.5	0.00	3.34	6.67	10.00	5.00
	5	0.00	6.67	13.34	16.67	9.17
	10	6.67	26.67	30.00	36.67	25.00
	20	10.00	36.67	43.34	53.34	35.83
Withania somnifera (L.) Dunal	2.5	0.00	6.67	10.00	13.34	7.50
	5	3.33	13.34	16.67	20.00	13.34
	10	6.67	16.67	20.00	23.34	16.67
	20	10.00	33.34	36.67	43.34	30.83
Salix mucronata Thunb.	2.5	0.00	0.00	0.00	3.34	0.83
	5	0.00	6.67	10.00	13.34	7.50
	10	6.67	13.34	16.67	20.00	14.17
	20	10.00	16.67	23.34	36.67	21.67
Lawsonia inermis L.	2.5	0.00	0.00	0.00	0.00	0.00
	5	0.00	0.00	0.00	3.34	0.835
	10	0.00	6.67	10.00	13.34	7.50
	20	6.67	13.34	20.00	33.34	18.33
LSD	(0.05)	4.01	8.93	9.45	12.51	8.81

mortality percentages were detected with all concentrations 1 day post-treatment then increased gradually.

Comparative toxicity of the tested materials on *Monacha obstructa* using three methods after 21 days of treatment under laboratory conditions

The data presented in Figure 1 compare the toxicity of plant extracts, *L. inermis*, *S. mucronata*, *S. nigrum* and *W. somnifera* with the different methods. The results indicated that, *S. nigrum* extract was the most toxic of all techniques, with LC_{50} values of 4.99, 14.79 and 23.00% for contact, leaf dipping and poison bait techniques, respectively. Also, the best technique was the residual film technique, as it gave the lowest LC_{50} value and the highest bioactivity compared to the other methods. It was also noted that the tested plant extracts exhibited the highest toxicity against the tested snail when applied using the thin-film method, compared to poison baits and leaf-dipping methods. Furthermore, this study showed that using plant extracts was more effective in controlling the terrestrial snail *M. obstructa* than using plant powders.

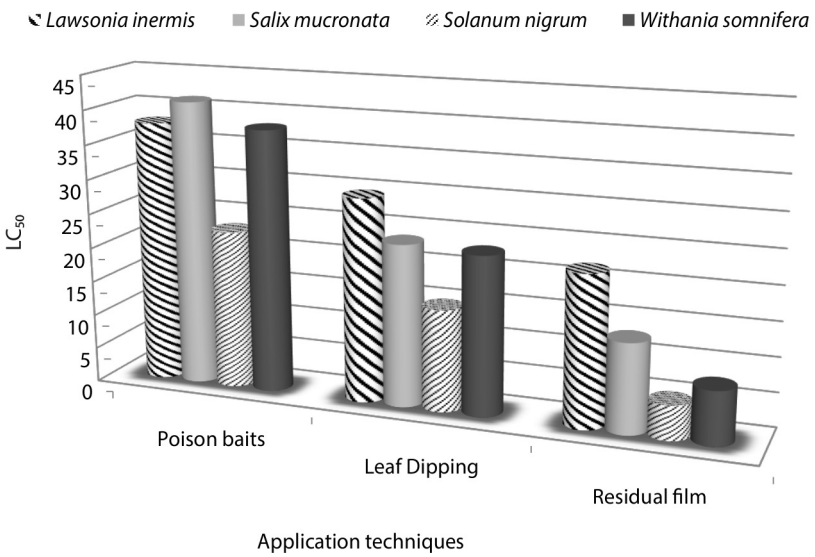

Fig. 1. Efficiency of different plant materials against *Monacha obstructa* by poison baits, leaf dipping and residual film, techniques after 21 days

Molluscicidal activity of certain plant extracts against *Monacha obstructa* land snails under field conditions in Sohag governorate

Two plant extracts were tested to control *M. obstructa* in citrus orchards using spray and in Egyptian clover fields using baits in Arab Sabha village, Sohag. Data in Table 5. showed that the reduction percentages of *M. obstructa* infesting orange and mandarin trees and Egyptian clover plant, treated with plant extracts *S. nigrum* and *W. somnifera* increased gradually with time. Twenty-one days post-treatment, reduction percentages of *S. nigrum* and *W. somnifera* were 65.55 and 39.72%, respectively, for the spray technique, while they were 54.35 and 40.29% for the bait technique. Also, the results indicated that *S. nigrum* extract was the most toxic.

Discussion

Plant extracts and natural powders are environmentally safe alternatives to the use of pesticides. They are used as one of the recent trends in integrated control programs. In our present study, it is obvious that there were variations in toxicity according to the type of plant extract, powder and method of application. These differences in toxicity levels may have been due to various classes of compounds responsible for molluscicidal activities of plants. Abdel Kader *et al.* (2007) found that using some plant water extracts with spraying was more efficient against land snails *Monacha cartusiana* (Müller) and *Theba pisana* (Müller) than using poisonous foods or grinding plant parts. Also, Ali *et al.* (2013) studied the use of the aqueous extract of *Solanum nigrum* as a molluscicide to control the brown garden snail, *Eobania vermiculata* (Müller) under laboratory conditions. The present study revealed that the rate of snail mortalities of the tested snails depended on the dose and duration of exposure to this extract. The highest mortality rates were noticed at doses of 8%. Large amounts of mucus were secreted by snails just after their soft bodies contacted the extract. Mourad (2014) studied molluscicidal effects of five ethanolic crude extracts, cumin (*Cuminum cyminum* L.), golden shower (*Cassia fistula* L.), umbrella tree (*Melia azedarach* L.), olive (*Olea europaea* L.) and pomegranate (*Punica granatum* L.) against land snail *Monacha obstructa* using three methods under laboratory conditions. The results indicated that the ethanol crude extract of cumin was the most toxic extract followed by golden shower, umbrella tree and pomegranate extracts while olive extract had the lowest effect. In addition, the contact technique of the tested plant extracts was the most effective method of application. The LC_{50} values of cumin, golden shower, umbrella tree, olive and pomegranate extracts when applied as contact were 250, 325, 635, 1500 and 910 ppm, respectively, for *M. obstructa* land snail. Abd El-Atti *et al.* (2019) used ethanolic ginger extract as a natural and environmentally safe molluscicide against *M. cartusiana*. They revealed that a low concentration (20%) of ethanolic ginger extract caused 10% mortality of clover snails after 1 day of exposure and 66.7% mortality after 28 days. The highest mortality percentage (90%) was recorded after 28 days of treatment with 40% ginger extract. Ali *et al.* (2020) studied the molluscicidal activity of three weed extracts namely primpernel (*Anagallis arvensis* L.), nightshade (*S. nigrum* L.) and nutsedge (*Cyperus longus* L.) against *M. obstructa* under laboratory conditions. Results indicated that chloroform and hexane extracts of primpernel appeared highly toxic when applied as baits on *M. obstructa* adults while nutsedge was found to have lower toxicity based on LC_{50} values. Also, chloroform extract of nightshade and hexane extract of primpernel were the most potent with contact, whereas nutsedge was the least effective. Regarding application, Ismail *et al.* (2011) examined the molluscicidal activity of clove powder against *M. cartusiana* snails as bait under field conditions. The results indicated that 40% concentration gave 62.4% population reduction after 21 days of treatment.

Table 5. Molluscicidal activity of certain plant extracts against *Monacha obstructa* land snails under field conditions at Sohag governorate

Plant extracts	Con.	Percent of population reduction after indicated days				Mean
		1	7	14	21	
Spray technique						
Solanum nigrum L.	20%	12.95	32.48	49.77	65.55	40.18
Withania somnifera (L.) Dunal		1.09	11.40	16.30	39.72	17.12
Bait technique						
Solanum nigrum L.	30%	34.50	49.83	21.40	54.35	40.00
Withania somnifera (L.) Dunal		2.48	17.85	26.03	40.29	21.66

Conclusions

In conclusion, the aqueous plant extracts of *S. nigrum* provided a natural and environmentally friendly alternative as a molluscicide for controlling the snail species *M. obstructa* and confirmed its possible use in integrated control programs to reduce the use of chemical pesticides. The accessibility and ease of preparation and application further enhance the viability of using these extracts, which makes it a subject of interest for further studies on the contents of this plant.

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