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EFFECT OF NEEM EXTRACT, NEEMAZAL T.S. ON TWO LAND SNAIL SPECIES UNDER LABORATORY CONDITIONS.

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ABSTRACT

The effect of neem extract (Neemazal T.S.) on the eggs hatchability and adult snails of two land snail species, *Monacha cartusiana* and *Eobania vermiculata* as well as its effect on food consumption of Juvenile snails *M. cartusiana* under laboratory conditions was also evaluated. Results revealed that the hatchability of the two tested snail eggs decreased to reach 50, 42.5% on the highest concentration (400 ppm.), respectively. Regarding the effect of neem extract (Neemazal T.S.) against adult snail *Monacha cartusiana* mortality percentages reached to 20, 40, 55% in the end of experiment for the three tested concentration 0.625, 1.250 and 2.5%, respectively. Neemazal T.S. exhibited antifeeding effect on the juvenile snails of *M. cartusiana*. Food consumption reached to 9.6 mg. (after 3 days) for the highest concentration (200 ppm.) compared with 31.9 mg. for untreated juveniles.

INTRODUCTION

Recently land snails had become one of economic serious pests in different governorates in Egypt. It is causing serious yield reduction of infested filed crops and fruits (Kassab and Daoud 1964 and Nakhla and Tadros 1995). Among these pests, the glassy clover snails, *Monacha cartusiana* (Muller) and the brown garden snail, *Eobonia vermiculata* (Muller) which considered the most predominant snails in all localities at Sharkia Governorate which attacking agronomic, horticulture and ornamental plants (El-Okda 1979; Ghamry *et al.* 1993; Ismail 1997 and Mahrous *et al.*2002). Though some molluscicides have been proved much effective in killing the snails, the use of these chemicals is not being encouraged nowadays due to environmental pollution. Therefore, considerable effort has been paid to control the pests through the use of natural products which derived from plant origin, as a potential source of pest control agent. Of these, extract from the neem tree, *Azadirachta indica* (A. Juss)(Meliaceacae) which native to India (Roxburgh, 1874). Azadirachtin (AZA), extracted from the seeds of the neem tree, is considered to be the most biologically active component of neem (Warthen, 1989), beside salannin, meliantriol and nimbin (Jacobson 1990). These natural pesticides are known to have strong toxicity effects against a wide spectrum of insects, including anti-feedant, growth regulatory, and sterility effects (Schmutterer *et al.* 1981; Jacobbson, 1989 Su and Mulla 1998a; Singh 2003) ; repellency effects (Dhiman and Sharma 1994) and inhibition of oviposition (Dhar *et al.* 1996), yet it has a low toxicity towards beneficial organisms such as parasitoids, predators and pollinators (Lowery and Isman 1995; Naumann and Isman 1996; Raguraman and Singh 1999), can degrade rapidly in the environment (Isman 1999) and safe to carabid bettles, the main predators of snails (Forster 1991; Mohapatra *et al.* 1991 ;

Srinivas and Madhumathi 2005). Although limited in number, some investigators studied the efficacy of extract of neem against different species of terrestrial snails. Among the former studies, those carried out by Ebenso (2004) that showed no effects on the controls or snails exposed to neem seed oil extract. Crude extract of bark, roots and leaves of neem at 500 and 700 mg kg (-1) produced mortality after exposure for 48 h for *Limicolaria aurora* and 72 h for *Achatina marginata*. Neem extract has also been found to possess significant activity against *Lymnaea acuminate* and *Indoplanorbis exustus* in binary and tertiary combinations with other molluscicidal plant products (Singh and Singh, 2001 & 2004). Shoaib *et al.* (2009) studied the toxicity of the commercial neem based insecticide Nimbecidine® on food consumption & egg hatchability of terrestrial snails . The aim of this study assess the efficacy of neem extract (Neemazal T.S.1%) on egg hatchability and food consumption of two land snails, *Monacha cartusiana* and *Eobania vermiculata* under laboratory conditions.

MATERIALS AND METHODS

1- Tested material:

The formulation of neem extract which used in this test is Neemazal T.S. 10000 ppm. This compound was obtained by Tri- Folia –M Gmbh-Company, Lahnay, Germany.

2- Collecting of snails:

Specimens of the terrestrial snail, *M. cartusiana* and *E. vermiculata* were collected from infested Egyption clover field at Zagazig district, Sharkia Governorate. Snails were put in rearing containers (50 × 30 × 30 cm.) and fed daily on fresh cabbage leaves. Egg layed was observed daily and all deposited egg clutches in the rearing containers were carefully collected for the ovicidal test.

3- Egg tests:

Eggs of *M. cartusiana* and *E. vermiculata* snails were obtained by the previous mentioned manner and clutches were removed carefully, put in Petri-dish, washed with distilled water and then prepared for the following test. Clay soil were taken from the upper 15 cm. surface layer of the farm at Hehia county, Sharkia Governorate. The samples were air dried, cleaned from plant parts, thoroughly mixed, crushed, sieved through 2 mm. sieve and subjected to application. Four concentrations from Neemazal T.S. were prepared i.e.50, 100, 200 and 400 ppm. Tested eggs were dipping for one minute in all concentrations. Sieved clay soil were put in plastic boxes (3/4 kg. capacity) and irrigated to reach the field capacity. Small holes were made by glassy needle and ten eggs were placed into it. Four replicates were used for each concentration and check control were prepared using soil free from any Neemazal extract. Soil was remoisted as required. The boxes were tightly covered with cloth netting secured with rubber band to prevent hatchlings from escaping, Boxes were examined daily and hatching percentages were recorded. Obtained data were subjected to statistical analysis using S.E.

4- Effect of Nemazal T.S. on *M. cartusiana* adult snails.

Tested animals of *M. cartusiana* snails were collected before treatment from some infested fields at Zagazig region, Sharkia Governorate. They were allowed to acclimatize under laboratory conditions for two weeks and fed on cabbage leaves. Three concentrations of Neemazal T.S. i.e. 0.625 %, 1.25% and 2.5 % were prepared by incorporating the appropriate amount of each concentration with bran bait to give 100 parts from poisonous baits. Four plastic boxes (3/4 k capacity) were used for each concentration. Control treatment was prepared using bran bait only. Five grams of each concentration were spread into each box. Ten adult snails were introduced in each box and then tightly covered with muslin clothes netting and secured with rubber band to prevent snails from escaping. Mortality percentages were recorded after 1, 7, 14 and 21 days post-treatment. Observation of mortality entailed using stainless steel needle according to El-Okda (1981). Dead snails were removed after testing and mortality percentages were calculated during experiment periods. Data were statistically analyzed to obtain L.S.D.

5- Food consumption experiment:

Food consumption by juveniles of *M. cartusiana* snails was tested when treated with Neemazal T.S. under laboratory conditions. Clay soil was prepared by the previous manner mentioned in egg tests. About 400 gms of soil were put in each plastic boxes (3/4 kg capacity) and irrigated to reach field capacity. Five juveniles (diameter 6-8 mm.) of *M. cartusiana* snails were introducted in every box. Three concentrations were prepared i.e. 50, 100 and 200 ppm from Neemazal T.S. Disc from cabbage were dipped for one minute and offered to the socials three treatment carried out and control treatment was conducted. Three replicates discs were weighted daily before and after application during 3 successive days. Consumed amount of food was measured daily according to Waldbauer (1964). Data were subjected to statistical analysis using L.S.D.

RESULTS AND DISCUSSION

1- Egg tests:

Data presented in Table (1) show the effect of Neemazal T.S. on hatchability of *M. cartusiana* and *Eobania vermiculata* eggs by dipping technique under laboratory conditions. The results show that the hatchability was decreased by increasing the concentration of Neemazal T.S. in two tested snail eggs. Hatchability was 77.5±6.3 and 70.0±4.1 for *M. cartusiana* and *E. vermiculata* snails at the lowest concentration (50 ppm), while it reach 50.0±4.1 and 42.5±4.7 at the highest concentration (400 ppm) for the two land snails eggs respectively. Aioub et al (2000) reported that carbosulfan (furadan 10% G., nematicide) and Tralkoxydin (grasp 10% E.C., herbicide) were the most effective against the two land snail eggs, *M. cartusiana* and *E. vermiculata* when applied as soil treatment and dipping technique, respectively.

Table (1): Effect of Neemazal T.S. on egg hatchability of *M. cartusiana* and *E. vermiculata* under laboratory conditions.

Neemazal T.S conc. (ppm.)	Egg hatchability (Mean ± SE) (%)	
	M. cartusiana	*E. vermiculata*
50	77.5±6.3	70.0±4.1
100	65.0±6.5	62.5±4.8
200	60.0±4.1	52.5±4.8
400	50.0±4.1	42.5±4.7
Control	97.5±2.5	95.0±2.9

Shoaib *et al.* (2009) found that LC_{50} of Nimbecidine ® (containing azadirachtin 0.03%) for the treated eggs was 2.18 ml/ L and eggs failed to hatch at concentration of 10 ml/L, which caused 100% mortality of eggs. It is necessary to mentioned that land snails laid its eggs in the upper surface layer of soil, so; we can reduce the number of hatchability of this harmful snails during the breeding season. Although further field studies were necessary to evaluated these products under field conditions.

2- Adult snail experiment:

The efficacy of Neemazal T.S. formulation were determined under laboratory conditions as poisonous-baits against *M. cartusiana* snail. Data in Table (2) revealed that mortality percentages increased with the increase of Neemazal concentration and (0.625%) failed to exhibited any molluscicidal activity against *M. cartusiana* adult snail at the third day post-treatment. From the seventh day of treatment, mortality percentage began to increase while it reached 2,5,15 and 17.5% for the three tested concentrations, respectively. Mortality percentages increased by time elapsing to reach 20, 40, and 55 % at the end of experiment (28 days) for the three tested concentrations, respectively. Generally, it could be reported that the highest tested concentrations (2.5%) had the most effect as poisonous baits against *M. cartusiana* snails under laboratory conditions.

Table (2): Effect of neemazal T.S on egg hatchability of *M. cartusiana* adult snails under laboratory conditions.

Neemazal T.S conc.%	Mortality percentage after indicated days						Mean
	1 day	3days	7days	14 days	21 days	28 days	
0.625	0	0	2.5	7.5	12.5	20.0	7.08
1.250	2.5	7.5	15	22.5	35.0	40.0	20.41
2.500	2.5	7.5	17.5	32.5	45.0	55.0	26.66
L.S.D.$_{0.05}$							ns

The literatures regarding molluscicide activity of neem extract on land snails are very lack.

Ebenso (2004) determined the effect of 350,500 and 700 mg/ kg of crude extracts of neem, *Azadirachta indica* (A Juss), on the edible land snail *Achatina marginata* and *Limicolaria aurora* (Jay). Crude extracts of bark, root and leaf of neem at 500 and 700 mg/kg produced mortality after exposure for 48 h for *L. aurora* and 72 h for *A. marginata*.

3. Food consumption:

Data in Table (3) show the effect of Neemazal T.S. on food consumption of *M. cartusiana* under laboratory conditions. Results revealed that Neemazal T.S. caused reduction in the food consumption of juveniles snails compared with untreated control treatment. Food consumption at the highest concentration after the third day post-treatment was observed reach the lowest values compared with control treatment where gave 9.6 and 31.9 mg/day for one juvenile snail, respectively. Regarding the mean of food consumption for the three tested concentrations, it gave 25.4, 23.8 12.9 and 32.9 mg. for 50, 100, 200 mg/juvenile and untreated control, respectively. Generally, it could be reported that Neemazal T.S. caused reduction of feeding of *M. cartusiana* snail juveniles. Singh and Singh (1998) reported that the mixture of cedar and neem oil were the most toxic to *Lymnaea acuminate* or garlic bulb powder was more toxic than the individual components.

Gabr *et al* (2006) reported that different neem preparation (Neemix4.5 ®) did show molluscicidal activity against *Monacha obtstructa* and *Eobania vermiculata* snails under laboratory and field conditions.

Table (3): Effect of neemazal T.S. on food consumptions (mg) of *M. Cartusiana* juvenile snails under laboratory conditions.

Neemazal T.S conc. (p.p.m.)	Food consumptions (mg)			
	1 day	2 days	3 days	Mean
50	24^{a}	30.1^{ab}	22.3^{a}	25.4
100	24^{a}	23.6^{cb}	24.0^{a}	23.8
200	18.2^{a}	10.9^{c}	9.6^{b}	12.9
Control	28.1^{a}	38.9^{a}	31.9^{a}	32.9
$L.S.D._{0.05}$	ns	15.311*	9.646**	

Shoaib *et al.*(2009) tested Nembicidine® on the effect of food consumption by *M. obstructa* snail under laboratory conditions. Results revealed that Nembicidine® has anti-feedant effect against *M. obstructa* reached 100% inhibition of feeding activity at the dose of 10 ml/ L.

On the other hand, Shoaib *et al* (2009) mentioned that Nembicidin® did not show ability to kill *Monacha obstructa* snail. It show a strong anti-feedant effect against the snail caused 100% inhibition of its feeding activity at a dose of 1 ml/L.

REFERENCES

Aioub, A.A.; Ismail, Sh. A.A. and Mohamdein, A.A.(2000). Toxicological and histological studies on some pesticides-treated land snails. Proc. of the first Inten. Con. On Biol. Sci. Fac. of Sci. Tanta Univ., 1(2) 19:38.

Dahr, R. ; Dawar, H.; Garg, S.; Basir, S.F. and Talwar, G.P.(1996). Effect of volatiles from neem and other natural products on gonotrophic cycle and oviposition of *Anopheles stephensi* and *A. culicifacies* (Diptera: Culicidae). J Med Entomol, 33(2): 195-201.

Dhiman, R.C. and Sharma V.P.(1994). Evaluation of neem oil as sandfly, *Phlebotomus papatasi* (Scopoli) repellent in an oriental sore endemic area in Rajasthan. Southeast Asian J Trop Med Public Health, 25:608-610.

Ebenso, I.E.(2004). Molluscicidal effect of neem (*Azadirachta indica*) extracts on edible tropical land snails. Pest. Mang. Sci. 60(2): 178-182.

El-Okda, M.M. (1979). Land snails of economic importance at Alexandria region with some notes on the morphological feature, classification, economic damage, and population on the ornamental plants. Agric. Res. Rev. 57 (1): 125-131.

El-Okda, M.M. (1981).Response of two land mollusca to certain insecticides. Bull. Ent. Soc. Egypt Econ. Ser. 12: 53-57.

Forster, P.(1991). EinfluB Von Pflanzenschutzmitteln auf-larven Und Adulte Von *Platymus dorsalis* (Pont) (Coleoptera: *Carabidae*) Und auf Adulte Von *Tachyporus hyphorum* (L) (Coleoptera: *Staphlinidae*) in Labor-Halbfreiland Und Freilandversuchen. Doctoral thesis, University of Giessen, Germany.

Gabr, W.M.; Youssef, A.S. and Khidr, F.K. (2006). Molluscicidal effect of certain compounds against two land snail species, *Monacha obstructa* and *Eobania vermiculata* under laboratory and field conditions. Egypt. J. Agric. Res., 84: 43-50.

Ghamry, E.M.; El-Deep, H.I. and Kokab, Y.A. (1993). Ecological and morphological studies on certain land snails at Sharkia Governorate. Egypt J, Appl. Sci. 8 (11):213 -225.

Ismail, Sh. A.A. (1997). Ecology, biology and control of certain terrestrial snails infesting some vegetables and field crops in Sharkia Governorate. Ph.D. Thesis. Fac. Agric. Zagazig Univ. 130 pp.

Isman, M.B. (1999). Neem and related natural products. In: Hall FR, Menn JJ (eds) Biopesticides: use and delivery. Human, Totowa.

Jacobson, M.(1989). Focus on Phytochemical pesticides. Vol.I, The Neem Tree. CRC press, Boca Raton, FL, p 197.

Jacobson, M. (1990). Review of neem research in the United States. In: Locke Jc, Lawson RH(eds)Proceedings of a workshop on neems, potential in pest management Programs. USDA-ARS,Beltsville, PP.4-14.

Kassab A. and Daoud H. (1964).Notes on the biology and control of land snails of economic importance in U.A.R. Agric. Res.,Cairo. 42:77-98.

Lowery D.T. and Isman M.B. (1995). Toxicity of neem to natural enemies of aphids. Phytoparasitica, 23: 297-306.

Mahrous, M.E.; Ibrahim, M.H. and Abdel-Aal, E.M.(2002). Ecological aspects of the glassy clover snail, *Monacha cartusiana* (Muller) under field conditions in Sharkia Governorate. Egypt. 2nd. Intern. Conf. PPRI. Cairo, Egypt, 21-24 December. Vol.1, 107- 114.

Mohapatra, R.N.; Patnaik, N.C.; Mishra, B.K. and Patnaik, H.P.(1991). Comparative toxicity of some insecticides and neem oil against brown plant hopper, *Nilaparvata lugens* (Stal) and its predators. Orissa J Agric Res, 4: 239-240.

Nakhla, J.M. and Tadros, A.W. (1995). Studies on the seasonal abundance of land snails on date palm shoots in Sharkia Govenorate. Egypt. J. Res., 73 (2): 347 – 355.

Naumann, K. and Isman M.B.(1996). Toxicity of neem (*Azadirachta indica* A. Juss) seed extracts to larval honeybees and estimation of dangers from field application. Am Bec J, 136: 518-520.

Raguraman, S. and Singh R.P. (1999). Biological effects of neem*(Azadirachta indica)* seed- oil on eggs parasitoid, *Trichogramma chilonis.* J Econ Entomol 92: 1274- 1280.

Roxburgh, W (1874). Description of Indian plants. Today and tomorrow, New Delhi, p 763.

Schmutterer, H; Ascher KRS and Rembold, H.(1981). Natural pesticides from The Neem Tree. Proc. 1st Int Neem Conf. Rottach-Egern , Germany, June 16-18, 1980. GTZ, Eschborn .

Shoaib, M.A.; Mahmoud, F.M; Loutfy, N; Tawfic, M.A. and Barta, M.(2009). Effect of botanical insecticide Nembecidine® on food consumption and egg hatchability of terrestrial snail *Monacha obstructa. J. Pest. Sci.* Accepted 30 June 2009. Springer–Verlag 2009.

Singh, S (2003). Effects of aqueous extract of neem seed kernel and azadiractin on the fecundity, fertility and post-embryonic development of the melon-fly, *Bactocera cucurbitae* and the oriental fruit-fly, *Bactocera dorsalis* (Diptera: Tephritidae). J Appl Entomol 127: 540-547.

Singh, K. and Singh, D.K. (1998). Molluscicidal activity of plant derived molluscicides. J. of herbs, Spices and Medic. Plant. 5(2): 67-72.

Singh, A. and Singh, D.K. (2001). Molluscicidal activity of *Lawsonia inermis* and its binary and tertiary combinations with other plant derived molluscicides. Indian J. Exp Biol, 39 (3): 263 – 268.

Singh, A. and Singh, D.K. (2004). Effect of herbal molluscicides and their combinations on the reproduction of the snail *Lymnaea acuminate.* Arch Environ Contam Toxicol, 46 (4): 470 – 477.

Srinivas, K. and Madhumathi, T.(2005). Effect of insecticide applications on the predator population in rice ecosystem of *Andhra pradesh.* Pest Manag Econ Zool, 13: 71-75.

Su, T. and Mulla M.S.(1998). Ovicidal activity of neem products(Azadirachtin) against *Culex tarsalis* and *Culex quinquefasciatus* (Diptera: Culicidae). J American Mosq Cont Assoc 14: 204-209.

Waldbauer, G. P. (1964). The consumption, digestion and utilization of solaneceous and non – solanaceous plants by larvae of the tobacco horn worm, *Procoparce sexta* (Johan.) (Lep. Sphingidae), Ent. Exp. And Appl. 7: 253 – 269.

Warthen Jr. J. (1989). Neem (*Azadirachta indica* A. Juss): Organisms affected and reference list update. Proc Entomol Soc Wash 91:367-388.

تأثير مستخلص النيم (نيمازال تى إس) ضد نوعين من القواقع الأرضيه معمليا

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أجريت هذه الدراسة بهدف تقييم فاعلية مستخلص النيم (نيمازال تى إس) كمبيد بيض ضد نوعين من القواقع الأرضيه هما قوقع البرسيم الزجاجى (موناكا كارتوسيانا) وقوع الحدائق البنى (أيوبانيا فيرمكيولاتا) وكذلك تأثير هذا المستخلص على الأفراد البالغة من قوقع موناكا كارتوسيانا بالإضافة إلى دراسة تأثير هذا المستخلص كمانع للتغذية على الأفراد الغير بالغة لقوقع البرسيم الزجاجى.

أوضحت النتائج أن لهذا المستخلص تأثير على حيوية البيض ومنع خروج الصغار منع حدوث الفقس حيث بلغت نسبة الفقس عند التركيز الأعلى (٤٠٠ جزء فى المليون) إلى ٥٠، ٤٢.٥% لكلا من قوقعى موناكا كارتوسيانا وإيوبانيا فيرمكيولاتا على التوالى.

بالنسبة لتأثير هذا المستخلص على الأفراد البالغة فقد وجد أن نسب الموت لقوقع موناكا كارتوسيانا وصلت إلى ٢٠، ٤٠، ٥٥% للتركيزات الثلاثة المختبره على التوالى. كما وجد أن لهذا المستخلص تأثير كمانع للتغذية للأفراد الغير بالغة لقوقع موناكا كارتوسيانا حيث أنخفضت كمية الغذاء المستهلك إلى (٩.٦ مجم) للفرد الواحد وذلك مقارنة بالمستهلك فى الأفراد الغير معاملة حيث وصلت إلى (٣١.٩ مجم) للفرد الواحد فى اليوم.

قام بتحكيم البحث

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