

## MOLLUSCICIDAL EFFICIENCY OF SOME PESTICIDES AGAINST *Helix aspersa* LAND SNAILS UNDER LABORATORY CONDITIONS

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### ABSTRACT

The relative toxicity of three pesticides namely, indoxacarb, lufenuron and *Bacillus thuringiensis* were determined in the laboratory against the brown garden snails (*Helix aspersa*). Methomyl involved as a common standard is being used recommended molluscicide against such pest. The toxicity index of the tested compounds was 100, 92.18, 88.05, 84.28 and 40.86 for methomyl, lufenuron, indoxacarb SC, *Bacillus thuringiensis* and indoxacarb EC, respectively. Also, data indicated that the percentages of mortality were increased by increasing the concentration of the tested pesticides. Lufenuron was the highest effect (63.33%) after 2 days while the lowest effect was indoxacarb EC (18.33 %) whereas indoxacarb SC and *B. thuringiensis* were intermediate. After seven days the results revealed that lufenuron and indoxacarb SC were the most effective against land snails (85.12%) and (78.44%) respectively, compared with methomyl (96.19%), while *B. thuringiensis* has a moderate effect (47.85%). Indoxacarb EC has a slight effect (32.14%); hence, most of the tested compounds used in the present study can be recommended for field – trials against *H. aspersa* in terms of efficacy.

### INTRODUCTION

Snails and slugs are among the most bothersome pests in many garden and landscape situations. The brown garden snail (*Helix aspersa*), is the common snail causing problems in California gardens; it was introduced from France during the 1850s for use as food. Terrestrial gastropods (molluscus: snails and slugs) are being abundantly distributed in north coast, new reclaimed lands and addition to delta region of Egypt Kassab and Daoud,(1964); El- Okda and Khalil,(1981) and Abo- Bakr(1997). Several species of slugs are frequently damaging, including the gray garden slug (*Deroceras reticulatum*), the banded slug (*Limax poirieri*), and the greenhouse slug (*Milax gagates*). Both snails and slugs are members of the mollusk phylum and are similar in structure and biology, except slugs lack the snail's external spiral shell, (Flint, 1998).. Snails and slugs feed on a variety of living plants as well as on decaying plant matter. On plants they chew irregular holes with smooth edges in leaves and can clip succulent plant parts. They can also chew fruit and young plant bark (Flint, 1998). Indoxacarb is the common name proposed for the S. isomer of oxadiazine derivative, which is the more insecticidally active isomer. This compound represents a new class of compounds has broad-spectrum insecticidally activity and yet be environmentally soft (Salagado, 1990).Lufenuron is a new insecticide effective on pyrethriod and organophosphates- resistant pests. It was soft on adult beneficial and predatory mites, safe on a wide range of crops and

suitable for integrated pest management (Harder *et al.* 1995). One of the most promising biocontrol approach that has received attention of many scientists is the development of *Bacillus thuringiensis* toxins as insecticides (Belfiore *et al.*, 1994). *B. thuringiensis* is one of the biological insecticide containing proteins that are highly toxic to insects. More than 182 species of insects have been found to be susceptible to *B. thuringiensis* based bioinsecticides (Dean, 1984).

Therefore, the present study was conducted in the laboratory to evaluate the relative toxicity of these new safe pesticides as molluscicides for use against land snails.

## **MATERIALS AND METHODS**

### **Tested Pesticides:-**

Suspension concentrate and emulsifiable concentrate formation of indoxacarb (Avanut<sup>®</sup> 15%): methyl (s)-N-[7.chloro -2,3,4,a,5-tetrahydro-4a (methoxy carbonyl) indeno [1,2-e] [1,3,4 ] oxadiazin - 2- yl carbonyl ] - 4? (trifloro methoxy) carbonilate; lufenuron ( Match<sup>®</sup> 5 %) : N-[[[ 2,5-dichloro-4-(1,1,2,3,3,3-hexafluoro-propoxy)-(phenyl)amino]carbonyl]-2,6-difluorobenzamide (CA); (Xen- Tan<sup>®</sup> 10.3%) *Bacillus thuringiensis*, aizawai and methomyl (Lannate<sup>®</sup> 90 %): S: Methyl-N-(methyl carbamoyl) oxyl - thioacetimidate .

### **Tested pests:-**

The brown garden snails *Helix aspersa* O.F. Müller were collected from vegetable fields in spring season 2004 from Koom - Hamada district El – Behera governorate. They were acclimatized for a week under laboratory conditions and fed on lettuce (*Lactuca sativa*) ad. lib.

### **Laboratory experiments:-**

#### **I – Toxicity studies**

Serial concentrations (2 , 4 , 6 , 8 , 1 , 12 and 15 X10<sup>4</sup> ppm) for each tested pesticide were prepared in aqueous solution, each concentration was replicated 3 times and the lettuce leaves were dipped in the aqueous pesticide preparation for 5 seconds then allowed to dry. Leaf discs were cut (2.5cm in diameter) and patches of 15 healthy animals were placed in it for seven days. Results were obtained after 48 hrs of treatment and the percentages of mortality were calculated and compared with the control group according to Abbott's formula (1925). Percentage of snails' mortality was plotted on log arithmetic – probability graph paper and LC<sub>50</sub> and LC<sub>95</sub> values could be estimated. The toxicity index was determined according to Sun (1950) as (A/B) X 100, where A and B are the LC<sub>50</sub> values of both the most toxic compound and the other tested compounds respectively. The obtained data were analyzed according to Litchfield and Wilcoxon method (1949).

**II –Biological effects:**

In this method, toxic wheat bran baits were prepared containing each LC<sub>50</sub> and 0.5 LC<sub>50</sub> values for each pesticides (w / w), the bait consisted of (wheat bran : molasses : blue dye) with the ratio 8:1:1 respectively (WHO, 1961). Bait were distributed in three replicates for each treatment, each replicate contain 10 snails and 10 grams of bait. Control was made using fresh water with the bait. Replicates were kept under laboratory conditions and suitable humidity required for snail activity.

Daily dead snails were counted and removed from each replicate. Mortality percentage was calculated and the molluscicidal potency was measured for all tested compounds after 7 days.

**RESULTS AND DISCUSSIONS**

Relative toxicity data of the four tested pesticides against the adult snails *Helix aspersa* are presented in table (1). Results indicated that methomyl was the most toxic compound against the adult land snail *Helix aspersa*. Whereas, indoxacarb EC was the lowest one. The tested compounds can be arranged in a descending order according to its LC<sub>50</sub> as follows: methomyl > lufenuron > indoxacarb SC > *B. thuringiensis* > indoxacarb EC. The difference between the LC<sub>50</sub> values of methomyl and that other tested pesticides was significant (p <0.05) as indicated by none overlapping of the respective 95% fiducial limits. The toxicity index showed that methomyl was about 1.084, 1.14, 1.18, and 2.45 times as toxic as lufenuron, indoxacarb SC, *B. thuringiensis* and indoxacarb EC, respectively.

**Table (1):- Toxicity of certain pesticides against land snails *Helix aspersa* following feeding on treated lettuce leaves for seven days.**

Compounds	LC <sub>50</sub> X 10 <sup>4</sup> ppm	95% fiducial limit x 10 <sup>4</sup> ppm		Slope ± S.E.	χ <sup>2</sup>	Toxicity index
		upper	lower			
Indoxacarb EC	14.5	19.4	9.6	2.18±0.14	5.90	40.68**
<i>B. thuringiensis</i>	7	9.6	4.3	1.12±0.96	3.90	84.28**
Indoxacarb SC	6.7	7.5	4.5	1.42±0.23	2.03	88.05*
lufenuron	6.4	6.7	5.3	1.16±0.15	2.92	92.18*
Methomyl	5.9	6.1	5.4	2.34±0.33	3.8	100

\* Significant difference at 0.05    \*\* Significant difference at 0.01

The data in table (2) indicate the toxic effect of the pesticides used on *Helix aspersa* after different time intervals. After 2 days, the lowest concentration (0.5 LC<sub>50</sub> values) of lufenuron, methmoyl, indoxacarb SC, *B. thuringiensis*, and indoxacarb EC caused 46.67, 32.50, 13.8, 10 and 3.67 mortality respectively. While the highest concentration (LC<sub>50</sub> values) showed that lufenuron had the highest effect (63.33%) followed by indoxacarb SC (62.67%), methmoyl was (53.33%), *B. thuringiensis* (20%) and the lowest effect was indoxacarb EC (18.33%) mortality. After 5 days, the mortality percent was increased with 0.5 or LC<sub>50</sub> respectively. After 7 days, lufenuron showed the highest effect (85.12%), while the lowest one was indoxacarb EC (32.14%).

Table (2): Efficacy of sub lethal concentration of the tested pesticides against land snails *Helix aspersa* using toxic bait at different time intervals.

No. of days	Con.	Indoxacarb EC		Indoxacarb SC		lufenuron		<i>B. thuringiensis</i>		Methomoyl	
		% Mortality	Lethal dose of active ingredient mg	% Mortality	Lethal dose of active ingredient mg	% Mortality	Lethal dose of active ingredient mg	% Mortality	Lethal dose of active ingredient mg	% Mortality	Lethal dose of active ingredient mg
1	A	9.33±1.5	11.4±0.14	51.00±2.14	35.5±0.12	56.67±1.12	3.7±0.20	10.00±1.11	7.21±0.01	53.33±2.13	22.0±0.11
	B	3.67±1.2	5.8±0.21	16.67±2.30	13.3±0.20	36.67±1.20	1.6±0.10	9.00±1.2	1.25±0.02	23.33±1.20	9.50±0.12
2	A	18.33±1.23	11.7±0.32	62.67±2.36	36.5±0.13	63.33±1.23	3.8±0.12	20.00±2.18	8.1±0.02	66.67±1.13	22.0±0.12
	B	3.67±1.23	6.1±0.22	16.67±2.30	13.8±0.15	46.67±1.50	1.7±0.22	10.00±1.20	1.35±0.01	32.50±1.15	10.6±0.15
3	A	20.00±2.12	14.0±0.32	66.33±3.14	37.3±0.33	76.33±2.13	3.8±0.11	31.10±2.39	8.1±0.10	76.3±2.17	22.1±0.12
	B	10.00±1.40	6.4±0.25	17.40±1.40	18.2±0.22	54.80±1.80	2.0±0.20	16.68±1.25	1.42±0.10	35.05±1.20	11.0±0.16
4	A	28.07±2.17	14.5±0.12	76.00±3.11	38.3±0.23	78.89±2.14	3.8±0.13	44.43±2.14	8.8±0.12	76.3±2.17	23.2±0.11
	B	24.07±1.6	6.5±0.20	34.43±1.60	18.5±0.25	65.67±1.60	2.1±0.15	24.43±1.50	2.35±0.12	38.40±2.40	11.2±0.60
5	A	30.18±1.13	15.5±0.25	76.00±2.22	39.4±0.25	83.03±3.12	3.9±0.20	44.86±2.01	9.1±0.20	80.65±1.18	24.4±0.14
	B	24.07±1.13	7.1±0.32	41.20±2.11	18.6±0.35	68.20±2.10	2.2±0.50	26.50±2.10	3.42±0.02	39.60±1.25	11.6±0.23
6	A	31.12±2.14	16.8±0.24	78.44±1.13	39.8±0.12	85.12±2.22	3.9±0.14	46.45±1.05	9.3±0.11	96.15±1.22	24.8±0.12
	B	25.50±1.24	7.4±0.33	45.3±2.20	19.0±0.18	68.50±1.20	2.3±0.17	26.5±1.05	4.2±0.12	42.50±2.30	12.2±0.24
7	A	32.14±2.13	17.3±0.26	79.13±1.36	41.9±0.14	85.12±1.56	4.1±0.11	47.85±3.00	9.3±0.03	96.19±2.17	24.8±0.12
	B	25.5±2.13	8.1±0.36	46.5±1.45	20.4±0.23	68.50±1.70	2.4±0.12	28.40±1.6	4.5±0.04	45.25±2.34	12.5±0.28
mean	A	24.16 ±2.33**	14.5±0.25	69.93±2.13*	38.4±0.18**	75.49±2.15	3.84±0.13	34.94±2.61**	8.55±0.11	77.93±2.13	23.33±0.12
	B	16.67±2.30**	6.77±0.24	31.16±1.25**	17.4±0.22	58.43±1.80**	2.04±0.11	20.21±1.8**	2.64±0.16	36.66±2.14	11.23±0.18

Mean % of mortality is the average of five replicates each one contain ten animals.

\* Significant difference at 0.05 \*\* Significant difference at 0.01

A = LC<sub>50</sub> B = 0.5 LC<sub>50</sub>

Comparing the toxic effects of the pesticides with 0.5 or LC<sub>50</sub> concentrations, it was found the highest effects were obtained by methomyl followed by lufenuron then indoxacarb (SC), *B. thuringiensis* and finally indoxacarb EC with significant differences among treatments as indicated in table (2).

Generally, lufenuron, indoxacarb SC and *B. thuringiensis* showed high molluscicidal effect against land snails, an important pest on most field crops, under laboratory conditions compared with methomyl itself. In addition, other positive properties of these compounds low toxicity to non- target organisms and short persistence in the environment. Salgado, (1990); Harder *et al*, (1996); and Pluschkell *et al.*, (1998).

These results agree with the findings by Hussien (2003) who indicated that methomyl was more effective as molluscicide compound using application and poisoned food techniques than indoxacarb. Kishore (1987) illustrated that the chemical control is the most powerful tool available for controlling snails. Zedan *et al* (1999) found that bacterial formulation was the most effective against land snails compared with Lannate.

## REFERENCES

- Abbot, W.S. (1925). A method computing the effectiveness of an insecticide. *J. Econ. Entom.*, 18:265-267.
- Abobakr, Y.A. (1997). Toxicological and environmental studies on some terrestrial gastropods. M.Sc. Thesis, Faculty of Agric.Univ. of Alexandria, Egypt.
- Belfore, G.J.; R.K. Vadlamudi; Y.A.Osman and L.A.Bula(1994). A specific binding protein from *Tenebrio molitor* for the insecticidal toxin of *Bacillus thuringiensis* subsp tenebrionis. *J.Bioch.and Biophys. Res.Comm.*200,(1):359-364.
- Dean, D.H.(1984). Biochemical genetics of the bacterial insect control agent0 *Bacillus thuringiensis* :Basic principles and prospects for genetic engineering. In:Kaplan,E.S.(Ed).*Biotech.and Gen.Eng. Rev.*341-363.
- El-Okda,M.M.K.and K.A. Khalil(1981). Land molluscaas economic pest attacking fruit orchards at Alexandria governorate. *Proc. 4<sup>th</sup> Arab Pesticides Conf. Tanta Univ.* 11:289-302.
- Flint ,L.(1998). *Pests of the Garden and Small Farm: A Grower's Guide to using less Pesticide.*, 2<sup>nd</sup> ed Oakland:Univ. Calf. Div. Agric. And Nat. Resources, Publication 3332.
- Harder, H.H.;S.L.Riley;S.F.Mocann and S.N.Irving(1995).Indoxacarb: a novel broad- Spectrum, environmentally soft, insect control compound. *Proc. Brighton Crop Protection Conf.Pests of diseases.* Brighton,UK, 449- 454.
- Harder, H.H.; S.L.Riley; S.F.Mocann and S.N.Irving(1996). Indoxacarb:a noval broad spectrum, environmentally soft, insect control compound. *Proc.Brighton. Crop Protection Conf. Pests of diseases* , Brighton, UK, 449-454.

- Hussien, A.A. (2003). Toxicological and Biochemical studies on some economic Pests. Ph.D. Thesis, Faculty of Agric. Univ. Alexandria, Egypt.
- Kassab, A. and H. Daoud (1964). Notes on the biology of land snails of economic importance in UAR. Agric. Rev. Cairo Min. of Agric. 42: 77-88.
- Litchfield, J.T. and F. Wilcoxon (1949). Simplified methods of evaluating dose effect Experiments. J. Pharmacol. and Exp. Therap. 96:99-113.
- Pluschkell, V.; A.R. Horowitz; G. Phyllis and I. Ishaaya (1998). DPX-MPO62 a potent compound for controlling the Egyptian cotton leafworm *Spodoptera littoralis* (Biosd). Pestic. Sci., 54:85-90.
- Salagado, V.L. (1990). Mode of action of insecticidal dihydro pyrazoles: selective block of impulse generation in sensory nerves. Pestic. Sci., 23:389-411.
- Sun, Y.P. (1950). Toxicity index: An improved method of comparing the relative Toxicity of insecticides. J. Econ. Entomol. 43:45-53.
- WHO (1961). Expert committee on bilharziasis. Wld. Hlth. Org. Techn. Rep. ser. 214-230.
- Zedan H.A.; A.A. Saleh and S.M. Abd-ail (1999). Bactericidal activity of *Bacillus Thuringiensis* against snails – toxicological and hostilgical studies. The 2<sup>nd</sup> Int. Conf of Pest Control, Mansoura, Egypt.

دراسات معملية لتقدير كفاءة بعض المبيدات على القواقع الأرضية  
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استهدفت هذه الدراسة معرفة كفاءة أربع مركبات حديثة آمنة على البيئة من مجاميع مختلفة هي مركب الماتش (لوفينبيرون ١٥%) والأفانت (إندوكساكارب ٥%) في صورتين مركز قابل للاستحلاب (EC) ومركز معلق قابل للانسياب (SC) والمبيد البكتيري الزنتاري (Bacillus thuringiensis ١٠,٣%) ومقارنتها بمركب اللانثيت (ميثوميل ٩٠%) الموصى به في مكافحة القواقع الأرضية. تم استخدام قواقع الحدائق البنية *Helix aspersa* المنتشر بصورة وبائية في محافظات الوجه البحري وذلك تحت الظروف المعملية. تم تقدير التركيز القاتل لنسبة الـ ٥٠% من الأفراد لكل المبيدات المستخدمة في التجربة. وأظهرت النتائج أن قيمة LC<sub>50</sub> لهذه المركبات كانت ٥,٤ و ٦,٤ و ٦,٧ و ١٤,٥ و ١٠ X جزء في المليون لمركب اللانثيت ثم الماتش ثم الأفانت في صورة المعلق قابل للانسياب ثم المركب البكتيري الزنتاري ثم مركب الأفانت في صورة مركز قابل للاستحلاب على التوالي. كما أوضحت النتائج أن مركب الماتش أعطى نتائج ممتازة في زيادة القواقع الأرضية تحت ظروف المعمل وصلت إلى نسبة زيادة ٧٥,٤٩% مقارنة بمركب اللانثيت ٧٧,٩٣% يليه مركب الأفانت في صورة SC بنسبة ٧٢,٧٩% ثم المركب البكتيري زنتاري بنسبة ٣٤,٩٤% كما أظهرت النتائج أن أعلى كفاءة لهذه المركبات كانت بعد سبعة أيام من المعاملة.