# HISTOLOGICAL CHANGES INDUCED IN THE MUCUS GLANDS OF BROWN GARDEN SNAIL *EOBANIA VERMICULATA* TREATED WITH MALATHION AND METHOMYL PESTICIDES.

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#### **Abstract**

The present study aimed to illustrate the histological effects of malathion and methomyl compounds on the mucous gland and the body wall of the land snail E. vermiculata at 0.5% level after 24 hours post-treatment. Mucus gland :- Tufted tubular and collection of mucous gland branches were formed by the effect of 0.5% malathion in mucous gland of E. vermiculata after 24 hours post treatment, at the same time, the quantity of mucous secreted was reduced and increased in volume of mucous cells. Also, effects of methomyl in mucous gland are cohesion of the branches of the mucous gland wide led to the desiccation of land snail and its death, and increase in the mucous cell volume with some change in the colour and it had been lonely and had no combination with each other . The body wall :- Histological effects of malathion on body wall of E. vermiculata led to presence of a large vacuoles spreading on the surface of skin wide, reducing the quantity of mucous secretion, while 0.5% methomyl showed no vacuoles on the skin surface wide.

**Key words:** Histological charges, Mucus glands, Brown garden snail, malathion and metnomy,

## INTRODUCTION

Land snails have become one of the most dangerous crop pests . They have direct bad effect resulting from feeding on plant and daily activity . Recently many agriculture processes helped spreading land snails in rural areas of Egypt .In the past they existed only in the Northern coast. On the other hand the rapid adaptation to the different places played an important role in this respect (Asran , 2001) . With the increasing numbers of land snails , the mechanical control has become an effective way to eliminate their effect. Moreover, it was necessary to use toxic chemical compounds to eliminate these pests. Several histological and biochemical studies were carried out to determine certain enzymes as well as the histological changes resulting from the toxic treatments to explain how much those land snails affected ( Fouad *et al* .2004 ). The present work was carried out to study some histological changes of 3

mucus gland and body wall of the land snail *Eobania vermiculata* treated by Methomyl and Malathion at adult stage.

# **MATERIALS AND METHODS**

#### 1-Tested animals:-

Adult snail of *E.vermiculata* were collected from different infested nurseries at Giza Governorate. Mean shell diameter of selected animals was <15mm . Healthy individual snails were kept separately in a small plastic boxes containing 8-10 cm moist optimal soil (clay, peat and sand 1:1:1) provided with fresh green lettuce leaves , covered with muslin secured with rubber band to prevent snails from escaping , and kept under the ideal laboratory conditions i.e. of  $20 \pm 2^{\circ}$ c and  $80 \pm 5^{\circ}$ R.H (Zidan *et al* .2001).

#### 2- Chemical insecticides used:-

- 1- Malathion 57.5% E .c, a phosphorus compound , at the concentration of 0.5~% . Chemical name : 0.0 -diamethyl S ( 1.2 dicarb ethoxy ethyl) phsporodithioate .
- 2- Methomyl 90%W.P ,a carbamate compound, at the concentration of 0.5% . Chemical name : S methyl N- { (methyl carbamoyl) oxy thioacetimidate }.

## 3-Chemical treatments:-

The two tested compounds were prepared just before application . Appropriate amount of each compound was diluted with water to obtain the required concentration as mentioned by Ghamry,1997. Fresh lettuce leaves were sprayed with the candidate compounds and were offered to the maintained land snail species after 24h starvation (Abd - El - Karim ,2000 ) . Five replicates were prepared for each treatment in addition to another one left with each treatment as a check control.(Fouad, 2002 ) . All groups of treated and untreated animals were examined for their mucous gland and body wall.

#### 4- Anatomical and histological studies :-

To observe the gross morphology of the soft parts of *E. vermiculata* the snails were anaesthetetized in 1% solution of chloral hydrate for 12 hours. The shell was then removed and the soft parts were fixed in 10 % formalin for at least 24 hour. Soft parts were progressively dissected with the help of binocular dissecting microscope. The mucous gland and the body wall of the treated animals were carefully removed photographed and compared with those of untreated (Shoieb,1997). Histological

preparation were inducted by light microscopy investigations , the soft parts of the snails were directly immersed in Bouin's solution for fixation which proved to be satisfactory for this work ( Cowie,1984). Small tissues representing different regions of both the mucous gland and body wall were washed , dehydrated , cleared and finally embedded in paraffin wax . Sections were cut 6 - 8 microns in thickness , mounted on glass slides and stained with haematoxylin and eosin . Photomicrographs (X40) were taken for both mucous gland and body wall and compared with those of untreated animals ( Mersal , 1990 ) .

# RESULTS AND DISCUSSION

Effects of malathion and methomyl pesticides on gland mucous and body wall of the land snail *Eobania vermiculata* after 24 hours post treatment compared with untreated snails are illustrated in Figs (1 - 9).

## A- Mucous gland:

**Normal untreated mucous gland :-** A pair of tufted tubular mucous gland are much branched and resemble little bunches of whitish sea – weed Fig (1). Also, The secretory cells forming the wall of the tubules are tall columnar cells containing the secretory granules Fig (2). The secretory cells are arranged around irregular Lumina,

Effects of malathion and methomyl pesticides on gland mucous:-Histological changes is mucous gland after 24 hours post treatment with 0.5 % malathion were shown in Fig (3) It can be seen clearly the complete adhesion of tufted tubular and wide reduce the quantity of mucous secreted to protect the land snail *E. vermiculata*. In addition , Fig (4) showed that, increase in the volume of cells and it is figure has been irregular ,while, effect of 0.5 % methomyl in mucous gland are illustrated in Fig (5) at 24 hours post treatment , cohesion and fusion of the branches of the mucous gland wide led to the desiccation of the land snail and it's death and increase in the cell volume of mucous gland cells with some change in the color and it had been lonely and had no combination with each other Fig (6).

Cook and Shirbhate (1983) noted that the nature of mucous secretion on the body surface varies, which suggests different functions for mucus on different areas of the epithelium. Luchtel *et al.* (1984, 1991) found that the secretions are often voluminous, and produced by cells of the epithelium, particularly by the mucous and channel cells.

# B-Body wall :-

The epithelium is composed primarily of a single layer of two types of columnar to cubical - shaped cells - micro villous cells and ciliated cells . These two cell types are distinguished by the presence of microvilli and cilia on their apical surfaces , respectively. Also ,the epithelium has large secretory cells ( vacuoles ) , which extend into the matrix of muscle and connective tissue that make up the sub epithelial part of the body wall .These large cells include mucous cells ,channel cells and calciferous cells Fig (7). Malathion compound induced histological changes on body wall of land snail E. vermiculata that illustrated in Fig (8) showed that led to a presence of a large vacuole in instead of presence of some small vacuoles spreading on the surface of skin wide reduce the quantity of mucous secreted to protect the animal from the effect of the pesticides so that led to a desiccation of the animal and it's death. Also, Fig (9) showed that, histological changes in body wall after 24 hours post treatment at 0.5 % methomyl, no vacuoles on the skin surface wide at 0.5% methomyl after 24 hours post treatment leading to never more secretion of the mucous so the faster death of land snail E. vermiculata . The results agree with Newell (1973,1977) who found that in terrestrial pulmonates, the skin is formed by an epithelium which is as well supplied with enzymes and mucous cells as the epithelium of vertebrate intestine. The skin of land snails contains special glands which secrete a covering of slime, which serves as a protection against desiccation. Also, the skin of land snails is importance as the structure which is exposed to contact with pesticides.

Deyrup – Olsen and Jindrova (1996) showed that the release of mucous is accompanied by the rupture of mucous granules which is triggered by ATP. The observed reduction of energy reserves (lipid and glycogen) in the crop and the hepatopancreas may be related to this phenomenon.

#### REFERENCES

- Abd El Karim, Nehmedo. 2000. Ecological and biological studies on some terrestrial African agricultural snail pests. Thesis of M .Sc .Of .African Studies Institute .Cairo , Univ , 123 -133 .
- Asran, Fawkyia, D. 2001. Evaluation and implementation of novel and environmentally safe approaches in I P M programme for terrestrial snails.

- Ph.D.thesis, Institute of Environ. Studies & Res., Ain Shams Univ ., Egypt , PP.199.
- Cook, A. and R. Shirbhate, 1983. The mucous producing glands and distribution of cilia of the pulmonate slug *limax pseudoffavues*. Journal of Zoology 201, 97 – 116.
- Cowie, R. 1984. The life cycle and productivity of the land snail *The pisana* (Mollusca: Helicidae). J.Anim. Ecol., 53: 311-326. Deyrup Olsen, I. and Jindrova, H. 1996. Product release by mucous granules of land slugs: *Ariolimax columbianus* as a model species. journal of Expen ental zoology, 276: 387 393
- Fouad, Maha , M . 2002. Effect of neem extract , Garlic acid and diflubenzuron on jaws morphology of some terrestrial molluscs . Egypt . j . Agric . Res., 80 (1) 155 – 167 .
- Fouad , Maha , M; Fatma . Khider , I . K . Ibrahim and A. M . A. Soliman . 2004. Laboratory studies on the molluscicidal effect of sumithion , Bindiocarb and Machet pesticides against three land snail species , J. Agric . Sci . Mansoura Univ ; 29 (1):451-455.
- Ghamry, E. M. 1997. Bioassay for two strains of bacteria Bacillus thuringiensis
  against certain land snails, under laboratory conditions. Egypt . J. Appl . Sci ., 12
  (5): 661-672.
- 8. Luchtel, D.L., Martin, A.W. and Deyrup-olsen. I. 1984 The channel cell of the terrestrial slug *Ariolimax columbianus*. Cell and Tissue Research 235, 143 151.
- Luchtel, D.L., Deyrup Olsen, I. and A.W. Martin. 1991 Ultrastructure and lysis of mucin containing granules in epidermal Pulmonata) Cell and Tissue Research 266,375 – 383.
- Mersal , H. T. I. 1990. Morphological, anatomical and histological studies on some land snails of agriculture importance in Egypt . M . Sc. Thesis , Faculty of Girls , importance in Egypt Ain Shams Univ : 1-199 .
- Newell , P.F. 1973. Etude de l'ulirastructure de l'epithelium dorsal et pedieux des limaces *Arion hortensis* Ferussac e \_*Agriolimax reticulatus* (Muller). Haliotis 3 , 131 -135. 1977. The structure and enzyme histochemistry of slug skin . Malacologia 16 ,183- 195.
- 12. Rita triebskorn, Kresten christensen and Ignaz heim 1998. Effects of orally and dermally applied metaldehyde on mucus cells of slugs (*Deroceras reticulatum*)

- depending on temperature and duration of exposure .J.Moll . Stud, 64 . 467-487.
- Shoieb , M.A. 1997. Ecological studies on some common Snail species and their control in Ismailia governorate. M. SC. Thesis, Fac. Agric ; Suez canal Univ :1-137 Egypt .
- 14. Zidan , Z. H ., H .I .El- Deeb, A.K.Sobeiha ,Setaita H. Ahmed, Fawkyia D. A. Asran and Maha Fouad. 2001. Molluscicidal and antifeedant effects of certain plant extracts against three land snail species in Egypt . j . Environ . Sci . Institute of Environmental Studies and Research Ain Shams University .Vol .2 , No.1, 57-74 .

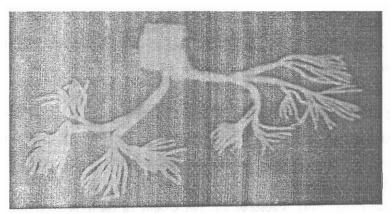


Fig. 1. Mucous gland of *E. vermiculata* in control group.

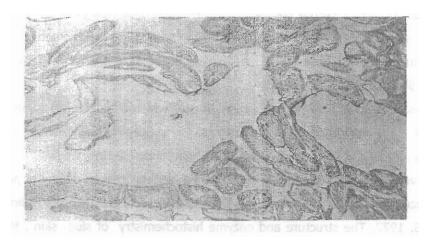


Fig. 2. Mucous cells of *E.vermiculata* in controal Group (x 40).

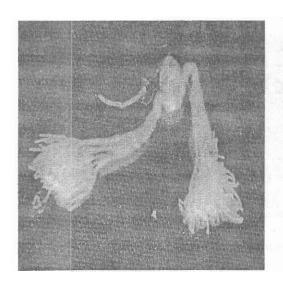


Fig. 5. Effect of methomyl on mucous gland of *E. vermiculata*.

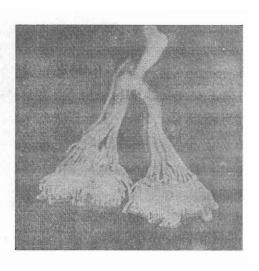


Fig. 3. Effect of malathion on mucous gland of *E. vermiculata*.

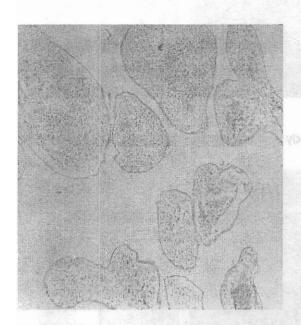


Fig. 6. Effect of methomyl on mucous cells of *E vermiculata* (x40).



Fig. 4. Effect of malathion on mucous cells of *E. vermiculata* (x40).

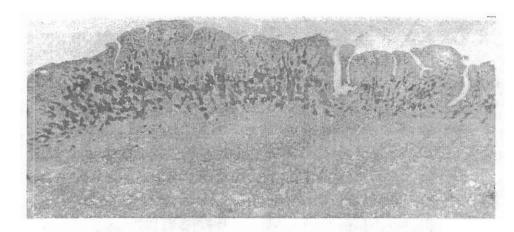


Fig. 7. Body wall of E.vermiculata in control group (x40).

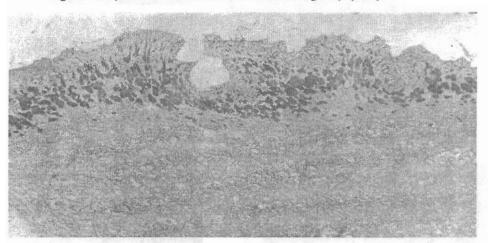


Fig. 8. Effect of malathion on body wall of *E.vermiculata* (x40).

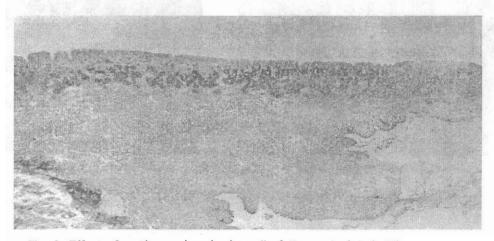


Fig. 9. Effect of methomyol on body wall of E.vermiculata (x40).

# التأثير الهستولوچي لمبيدي الملاثيون والميثوميل على الغدة المخاطية وخلاياها وجدار الجسم لقوقع الحدائق الكبير.

# مها فؤاد محمود

معهد بحوث وقاية النباتات – مركز البحوث الزراعية – وزارة الزراعة

تمت المقارنة بين تأثير مبيد كارباماتي (الميثوميل) ومبيد فوسفوري (الملاثيون) على التغيرات الهستولوجية لكل من الغدة المخاطية والخلايا المخاطية وجدار الجسم لقوقع الحدائق الكبيسر السيدين المعاملة بتركيز ٥٠٠%من كلا المبيدين وكانت النتائج المتحصل عليها كالآتي: - الغدة والخلايا المخاطية : - تتميز الغدة المخاطية بكثرة الفروع المفرزه للمخاطي فحدث انضمام لهذه الفروع في حاله المبيد الفوسفوري وكانت ملتصقة تماما في حاله المبيد الكرباماتي مما قلل من كميه المخاط المفر زه التي هي أساس حياه القواقع حيث يعتبس المخاط وسيله الحماية الأولى للقوقع . أما بالنسبة للخلايا المخاطية فحدث تضخم وانفصال لهذه الخلايا مع تغير اللون مما أدي إلى عدم قدرتها على إفراز المادة المخاطية وأدي ذلك إلى جفاف القوقع وموته وكان التضخم أكبر ما يمكن مع المبيد الكرباماتي (الميثوميل) عنها في حاله المبيد الفوسفوري (الملاثيون) .

- جدار الجسسم: - يحتوي جدار الجسم في حاله القواقع الغير مصابه على فراغات أو جيوب منتشرة على جميع أجزاء جدار الجسم يمر من خلالها الإفراز المخاطي إلى خارج الجسم للحمايسة فعمل المبيد الكرباماتي على اختفاء هذه الفتحات تماما أما تأثير المبيد الفوسفوري فكان أقل حده حيث لوحظ تجمع معظم هذه الفراغات و أحدث فراغا كبير مما أدي إلى خلل في توزيسع إفسراز المسادة المخاطية على سطح الجسم وبالتالي جفاف الحيوان وموته .