EFFECT OF SOME PLANT EXTRACT ON *Melanoides tuberculatus* (Müller, 1774) and *Biomphalaria alexandrina* (Ehrenberg,1831) SNAILS IN LIBYA

Farg. S. El Serite[1] and Hatel El-kamali[2]

1. Faculty of Medical Technology, Missurata , Libya.
2. Islamic Omdurman University , Faculty of . Science , Al khartoum, Sudan

ABSTRACT:

The molluscicidal activity of different extracts of six plant species of plants belonging to two families were evaluated against two types of snails , *Melanoides tuberculatus* and *Biomphalaria alexandrina* in agricultural area of Taourgha region in Libya. The results clearly showed that all tested extracts were effective against the two snail types at different concentrations. The most potent one was found in methanol extract of *Euphorbia terracina* (methanol extract was L c 50 ppm/ml then *Nerium oleander* was L c 50 ppm/ml .

Key words: Molluscicidal activity, Plant extracts, *Melonides tuberculata, Biomphalaria alexandrina, Euphorbia terracina, Nerium oleander.*

INTRODUCTION:

Human schistosomiasis is a parasitic disease caused by digenetic trematode species of the genus *Schistosoma* which co-habitate the venous plexuses of the mammalian viscera. Schistosomiasis transmitted by freshwater gastropod molluscs which serve as intermediate hosts[(10-18)] .In the tropics and subtropics Schistosomiasis is the second most important parasitic disease after malaria in terms of prevalence, public health and socio-economic importance [(9-19)]. *Melanoides tuberculata* and *Biophalaria alexandrina* were found in large number in small ditches and several minor canals at small depth and were scanty in big canals. They preferred muddy water and usually swam on the surface clinging to the grass or aquatic plants. *B. alexandrina* serves as an intermediate host for *Schistosoma mansoni* , human acquires infection by contact with freshwater snails infested with *Schistosoma* cercariae, which actively penetrate his intact skin [(5-7)] and subsequently develop to the adult worms. Cercariae released into water by the infected snails, in which the parasite undergoes asexual larval multiplication. The snails in turn become infected by miracidia released from *Schistosoma* eggs, which reach freshwater with human faeces [(3-4-12)]. *M. tuberculata* is a species of freshwater snail with an operculum and parthenogenetic. This species is native to subtropical and tropical northern Africa and southern Asia. It is known to carry certain trematode parasites which can be dangerous to humans. These snails serve as first intermediate host for many trematodes . It was found that many plants are growning in Tourgha region like *Lantana camara, Nerium oleader, Ricinus comminus, Euphorbia terracina, Chrozophora tinctoria* and *Hyoscyamus albus* which have may biological activities as antimicrobial, treatment of malaria, rheumatism, and skin rashes[(6-1)(17)].

The aim of the present work is increased attention for the use of new molluscicides which are highly effective, rapidly biodegradable, less toxic ,readily available and easily applicable than synthetic molluscicides. Plant

molluscicides could be appropriate for snails, especially in developing countries [13-20].

MATERIALS AND METHODS:

Study Area: This study is conducted in agricultural Taourgha region at 240km from east of Tripoli and 38km from east of Missurata city.

Plant Materials: Whole plant materials of six species collected during October/November 2009. The plants were identified at the Department of Botany, faculty of science, Tripoli University [16]

Preparation of Extracts: 120 grams of each coarsely powdered plant leaves is macerated in 300-400ml of methanol (95%) and acetone for 24hours at room temperature (26-29^{o}C). The extract was filtered and the solvents were evaporated under reduced pressure by using Rotary evaporator at 40^{o}C.Serial dilutions were prepared from each extract to which snails are exposed i.e. 1000, 850,700, 500, 300, 150 and 75ppm [7]

Habitat and collection of snails:

Melanoides tuberculata (Fig.2) and *Biomphalaria alexandrina* (Fig. 1) are explained by that these two snails were found in large number in small ditches and several minor canals at small depth and were scanty in big canals. They preferred muddy water and usually swam on the surface clinging to the grass or aquatic plants .

The snails were collected by the method recommended by Mandahi Barth (1962). Identification of the collected snails was based upon morphological characters according to Mandahi Barth (1962.

*Melanoides tuberculata and Biomphalaria alexandrina*which is the intermediate host for *Schistosoma mansoni*. They are examined in the laboratory for patent trematode infections by being placed in glass beakers with clean water, leaves of lettuce, some stones and pump of air, then kepet the laboratory for a period up to four weeks and rescreened again(at the end of this period we found that the diameter of snails increased from 0.4cm to1.2cm). Only those snails free from any infection and measuring 8-10mm in diameter are used in the laboratory experiments.

Ten snails of each type were placed in metanol in beaker containing 50ml of each concentration. Ten snails are put in a separate beaker containing 50ml of distilled water as a control. After 24 hours, snails were transferred into beakers containing 50ml of distilled water for recovery. They were examined after 24 hours for noting the dead as well as live ones. A snail is confirmed dead if it was remained immobile after having been observed for five minutes with the aid of 10 magnification hand lens and either retracted well into or hanged out of the shell, with the body and shell discoloured. Each experiment was repeated three times [7]

METHODS OF ANALYSIS

Probit regression analysis (SPSS/inc) was carried out for all tested plants to determine the LC_{50} and LC_{90} values. The slope of the regression line was used to assess the effect of the extract.

RESULTS AND DISCUSSION:

Table (1): Mortality rates among *M.tuberculata* and *B.alexandrina* snails (N=10) exposed to different concentrations of methanol extract of *Euphorbia terracina*

Dose of extract in (ppm)	*Melanoides tuberculata*				*Biomphlaria alexandrina*			
	Mortality %				Mortality %			
	Exp.1	Exp.2	Exp.3	mean	Exp.1	Exp.2	Exp.3	mean
75	70	60	70	66.6	70	80	70	73.3
150	70	80	80	76.6	80	80	90	83.3
300	100	100	80	100	100	90	100	96.6
500	100	100	100	93.3	100	100	100	100
700	100	100	100	100	100	100	100	100
850	100	100	100	100	100	100	100	100
1000	100	100	100	100	100	100	100	100

Table (2): Mortality rates among *M.tuberculata* and *B.alexandrina* snails (N=10) exposed to different concentrations of acetone extract of *Euphorbia terracina*

Dose of extract in (ppm)	*Melanoides tuberculata*				*Biomphlaria alexandrina*			
	Mortality %				Mortality %			
	Exp.1	Exp.2	Exp.3	mean	Exp.1	Exp.2	Exp.3	mean
75	20	20	20	20	30	20	30	26.6
150	10	20	10	13.3	10	30	20	20
300	20	10	20	16.6	20	20	10	16.6
500	30	20	20	23.3	20	30	30	26.6
700	90	100	90	93.3	90	100	100	96.6
850	100	100	100	100	100	100	100	100
1000	100	100	100	100	100	100	100	100

Table (3): Mortality rates among *M.tuberculata* and *B.alexandrina* snails (N=10) exposed to different concentrations of metanol extract of *Nerium oleander*

Dose of extract in (ppm)	*Melanoides tuberculata*				*Biomphlaria alexandrina*			
	Mortality %				Mortality %			
	Exp.1	Exp.2	Exp.3	mean	Exp.1	Exp.2	Exp.3	mean
75	10	20	10	13.3	0.0	10	0.2	3.3
150	70	60	70	73.3	50	60	50	53.3
300	90	80	80	76.6	70	50	60	60
500	100	100	100	100	100	100	100	100
700	100	100	100	100	100	100	100	100
850	100	100	100	100	100	100	100	100
1000	100	100	100	100	100	100	100	100

Table (4): Mortality rates among *M.tuberculata* and *B.alexandrina* snails (N=10) exposed to different concentrations of acetone extract of *Nerium oleander*

Dose of extract in (ppm)	*Melanoides tuberculata*				*Biomphlaria alexandrina*			
	Mortality %				Mortality %			
	Exp.1	Exp.2	Exp.3	mean	Exp.1	Exp.2	Exp.3	mean
75	40	50	40	43.3	50	40	50	46.6
150	60	70	60	63.3	70	60	60	63.3
300	80	90	90	86.6	90	80	80	83.3
500	100	100	100	100	90	100	90	93.3
700	100	100	100	100	100	100	100	100
850	100	100	100	100	100	100	100	100
1000	100	100	100	100	100	100	100	100

Table (5) Toxicity of the *Nerium oleander* methanol extract against the snail *Biomphalaria alexandrina*

Concentration	Mortality (%)	Log conc (X)	Probit (Y)
75	10	1.875	3.72
150	53.3	2.176	5.08
300	60	2.477	5.25
500	100	2.699	7.31

b=4.489556 LC_{50} = 1.269 ppm LC_{90} = 178.66 ppm

Table (6) Toxicity of the *Nerium oleander* acetone extract against the snail *Biomphalaria alexandrina*

Concentration	Mortality (%)	Log conc (X)	Probit (Y)
75	46.7	1.875	4.90
150	63.3	2.176	5.33
300	83.3	2.477	5.95
500	93.3	2.699	6.48
700	100	2.845	7.31

b=3.68873 LC_{50} = 100 ppm LC_{90} = 287.74 ppm

Table (7). Toxicity of the *Euphorbia terracina* methanol ext1ract against the snail *Melanoides tuberculatum*

Conc1entration	Mortality (%)	Log conc (X)	Probit (Y)
75	66.7	1.875	5.41
150	76.7	2.176	5.71
300	96.7	2.477	6.75
500	93.3	2.699	6.78
700	100	2.845	7.33

b=2.1011 LC_{50} = 156.234 ppm LC_{90} = 2143.22 ppm

T1able (8): Toxicity of the *Euphorbia terracina* acetone extract against the snail *Melanoides tuberculatum*

Concentration	Mortality (%)	Log conc (X)	Probit (Y)
751	20	1.875	4.16
1501	13.3	2.176	3.87
3001	16.7	2.477	4.01
5001	23.3	2.699	4.26
7001	93.3	2.845	6.48
8501	100	2.929	7.33

b=2.1709 LC_{50} = 111311.889 ppm LC_{90} = 924.698 ppm

Molluscicidal activity of *Nerium oleander* :

The comparative susceptibility of the snails: *M. tuberculata*, *B. alexandrina* to the action of different extracts (methanol, acetone,) from *N. oleander* has been determined.

Methanol extract:

The effect of various concentrations of methanol extract of *N. oleander* on adults of *M. tuberculata* and *B. alexandrina* snails after 24 hours exposure are listed in (Table 1) . The results of mortality were statistically analyzed using Probit analysis (spss/inc) The LC_{50} and methanol extract of M. tuberculata LC_{90} of this extract against *M. tuberculata* were 1.3 and 256.6 ppm respectively. The LC_{50} and LC_{90} of the same extract against *B. alexandrina* were 1.269 and 178.7 respectively . There was a difference between molluscicidal activities of methanol extract of *N. oleander* against two tested snails. *M. tuberculata* were more sensitive to *N. oleander* extract than B. *tuberculata*.

Acetone extract:

The effect of various concentrations of acetone extract of *N. oleander* on adults of *M. tuberculata*, *B. alexandrina* snails after 24 hours exposure are listed in (Table 6). The results of mortality were statistically analyzed using Probit analysis. The LC_{50} and LC_{90} of this extract against *M. tuberculata* were 103.3 and 229.0 ppm respectively. The LC_{50} and LC_{90} of the same extract against *B. alexandrina* were 100 and 287.7 ppm respectively.

Molluscicidal activity of *Euphorbia terracina* :

The comparative susceptibility of the snails: *M. tuberculata*, *B. alexandrina* to the action of different extracts (methanol, acetone) from *E. terracina* has been determined.

Methanol extract:

The effect of various concentrations of methanol extract of *E. terracina* on adults of *M. tuberculata*, *B. alexandrina* snails after 24 hours exposure are listed in Table 1. The results of mortality were statistically analyzed using Probit analysis spss/inc. The LC_{50} and LC_{90} of this extract against *M. tuberculata* were 56.2 and 243.22 ppm respectively. The LC_{50} and LC_{90} of the same extract against *B. alexandrina* were 44.2 and 176.2 ppm respectively.

Acetone extract:

The effect of various concentrations of acetone extract of *E. terracina* on adults of *M. tuberculata*, *B. alexandrina* snails after 24 hours exposure are listed in Table. The results of mortality were statistically analyzed using Probit analysis. The LC_{50} and LC_{90} of this extract against *M. tuberculata* were 313.9 and 924.7 ppm respectively. The LC_{50} and LC_{90} of the same extract against *B. alexandrina* were 273.5 and 857.0 ppm respectively.

The toxicity values of methanol extracts of different plants are arranged in a decreasing order as follows *Nerium oleander* and *Eupnorbia terracina* and the toxicity values of the acetone extracts of two plants are arranged in adecreasing order as follows *Nerium oleander*.

The molluscicidal activity of most the tested plants extracts is probably due to the presence of alkaloide, flavonids terpenoids as well as phorble esters in E. *terracina* possess molluscicidal, properties [2) (15-14-11).]

In the potential molluscicides derived from local plants have attracted the attention due to high costs of imported symthetic molluscicides.

Treatment of *Schistosoma* and *Fashiola* infections remains highly problematic. In schistosomiasis, praziquantel is faced with failure to prevent reinfection as a

result of development of drug resistance schistosoma strain and serious side effects, Treatment of *Fasciola* requires high or multiple doses of drug with frequent side effects.(15)

Fig. 1

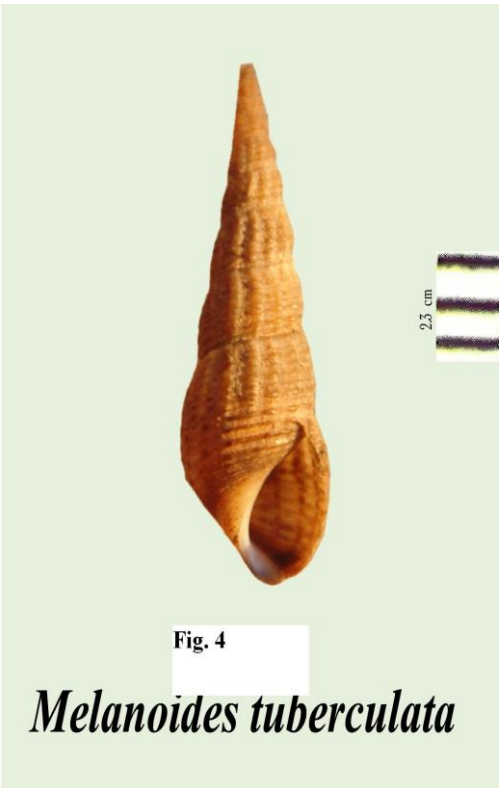

Fig.2

REFERENCES:

1. **Abdalla M. Elimam, Khitma H. Elmalik and Faysal S. Ali (2009)**. Larvicidal, adult emergence inhibition and oviposition deterrent effects of foliage extract from *Ricinus communis*L. against *Anopheles arabiensis* and *Culex quinque fasciatus*in Sudan. Tropical Biomedicine 26(2): 130–139.
2. **Abdel Gawad M, Anwar F., Refahy L. Hamed M. and EL Amin S. (2000)**. Structure elucidation of the separated contents of Dature a nnoxia mollus cicidal activity, Egypt. J. Biomed Sci. 6: 150-152-8.
3. **Benson, A. J. (2008).** *Melanoides tuberculatus*. USGS Nonindi genous Aquatic Species Database, Gainesville.
4. **Brown, D.S. (1994).** Freshwater Snails of Africa and their Medical Importance. 2nd ed., Taylor and Francis Limited, London.
5. **El Ridi, R. (2002).** Toward a complete immunity-inducing vaccine for Schistosomiasis. J. Parasitol.**88(5):** 1049-1050.
6. **Erlânio O. Sousa, Natálya F. Silva, Fabiola F.G. Rodrigues, Adriana R. Campos, Sidney G. Lima, and José Galberto M. Costa (2010)**. Chemical composition and resistance-modifying effect of the essential oil of *Lantana camara* Pharmacogn Mag.; 6(22): 79–82.

7. **Fayez A. B. (2009)**. Use of some plant extract to control *Biomphalaria alexandrina*snails with emphasis on some biological effects. World Appl. Sci. J., 6(10),1335-45.
8. http://nas.er.usgs.gov/queries/FactSheet.asp?speciesID=1037**.**
9. **James, S. and Colley, D. (1995).** Schistosomiasis. Curr. Opin. Infect. Dis.**8:** 351-355.
10. **Lockyer, A.E., Olson, P.D., Ostergaard, P., Rollinson, D., Johnston, D.A., Attwood, S.W., Southgate, V.R., Horak, P., Snyder, S.D., Le, T.H., Agatsuma, T., McManus, D.P., Carmichael, A.C., Naem, S. and Littlewood, D.T.J. (2003).** The phylogeny of the Schistosomatidae based on three genes with emphasis on the inter relationships of *Schistosoma* Weinland, 1858. Parasitol.**126:** 203-224.
11- **Mandahl-Barth, G. (1962).** key to the identification of East and Central African freshwater snails of medical and veterinary importance. Bull. Wld. Health Org., 27(1): 135-150.
12. **Mohamed MR, Refaie LA (2001).** Molliscicidal, parasitological and toxico-logical studies on Lantan camara plant . J. Egypt pharmacol Exp. Ther , 20: 265-286.
13. **Nakano, E., Watanabe, L.C., Ohlweiler, F.P., Pereira, C.A. and Kawano, T. (2003).** Establishment of the dominant lethal test in the freshwater mollusk *Biomphalaria glabrata* (Say, 1818). *Mutation Res.***536:**145-154.
14. **Osman, E.A.; Mohammed, E.M.; Abu Elreesh, B.I. and Elegami, A.A. (2007)**. Molluscicidal activity of *Combretum glutinosum*. *Int*. J. mol. and adv. sci., 3(4),151-54.
15. **Qing -Wen Shi, Hiao-HuiSu and Hiromosa Kiyota (2008)**. Chemical and pharmacology research of the plants in Genus Euphorbia, chem.. Rev. 108, 4295-4327.
16. **Refahy L.A. (2002).** Biological and chemical studies on *Peganum harmala* (Zygophyllaceae) Egypt. Schistosomiasis infect. Endem , Dis 24: 69-79.
17. **Samir A. M. Abdelgaleil and Mohamed E. I. Badawy (2006).** Acaricidal and Molluscicidal potential of three essential Oilsisolated from Egyptian plants. PP 213.
18. **Sharma S. , Singh T. and Vijayvergia R. (2009)**. Molluscicidal activity of some medicinal plants . Journal of Herbal Medicine and Toxicology,3 (2), 155-157.
19. **Smith, J.H. and Christie, J.D. (1989).** The pathology of human *Schistosoma haematobium* infections. In:Miller, M.J. and Love, E.J. (eds.). Parasitic Diseases: Treatment and Control. CRC Press, Boca-Raton, Florida. p. 3-17.
20. **Steinmann, P., Keiser, J., Bos, R., Tanner, M. and Utzinger, J. (2006).** Schistosomiasis and water resources development: systematic review, meta-analysis and estimates of people at risk. Lancet Infect. Dis.**6:** 411-425.
21. **Sushma S. and Singh D.K. (1998)**. Molluscicidal activity of *Neriumindicum* bark. Pp 233.

دراسة عن تأثير المستخلص الكحولي والاسيتوني لنباتي الدفلة واللبينة على قوقعي *Melonides tuberculata Biomphalaria alexandrina*

فرج سليمان السريتي[١] ، هاطل هاشم الكمالي[٢]
١- كلية التقنية الطبية مصراتة- ليبيا
٢- كلية العلوم جامعة أم درمان الإسلامية- الخرطوم- السودان

أظهرت النتائج أن نبات الدفلة أعطى جرعات قاتلة لقواقع ميلانوديس وبيومفالاريا LC_{50} وحسب التحليل الاحصائي في المستخلص الكحولي كانت تساويppm ١.٢٦٩ . وعند المستخلص الاسيتوني لنفس النبات كانت الجرعات القاتلة وحسب التحليل الاحصائي تساوى LC_{50}=103.2ppm LC_{90} = 229.01ppm ، أما نبات اللبينة وعلى نفس القوقعين السابقين فكانت جرعته القاتلة وحسب المستخلص الكحولي LC_{50}=156.2 ppm ، LC_{90} = 2143.2ppm ، أما المستخلص الاسيتوني لنفس النبات فكانت الجرعات القاتلة لكلا القوقعين كانت تساوى ppm. 8111311= LC_{50} ، LC_{90}=924.6ppm ،

التوصيات:

١- نوصي بضرورة الاهتمام بالنباتات الطبية ونوصي بضرورة حصرها وتصنيفها .
٢- إنشاء مركز يهتم بالنباتات الطبية العطرية .
٣- الاهتمام بالأبحاث في مجال النباتات الطبية والعطرية.
٤- مواصلة البحث العلمي في مجال تقنيات استخدام المستخلصات النباتية بجميع أنواعها.
٥- العمل على تطبيق نتائج الأبحاث في المجالات المختلفة.
٦- الاتجاه إلى المكافحة البيولوجية كبديل للمكافحة الكيماوية في مقاومة الآفات والحشرات الضارة وكذلك القواقع الناقلة للأمراض.
٧- التعاون العلمي بين الدول وخاصة العربية منها في هذا المجال البحثي كمصر والسودان والعراق على سبيل المثال وكذلك الدول ذات التقنيات العالية في المجال البيولوجي .