Evaluation of Euphorbia Aphylla, Ziziphus Spina-Christi and Enterolobium Contortisiliquum as Molluscicidal Agents

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Abstract: The present work was carried out to evaluate the molluscicidal activities of ethanoic extract of three medicinal plant species namely *Euphorbia aphylla, Ziziphus spina chriti,* and *Enterolobium contortisiliquum* against *Biomophalaria alexandrina* and *Lymnaea cailliaudi* (*nalatensis*) snails the intermediate hosts of schistosomiasis and fascioliasis respectively.the experiments were conducted in accordance with WHO guidelines. Probit analysis was used to determine the $LC_{50}$and LC_{90} after 24 hours exposure. The highest molluscicidal potency was recorded for *E. aphylla*. It exhibited significant molluscicidal activity on both snails' species. The LC_{50} and LC_{90} of this extract against *Lymnaea cailliaudi* were 0.66 and 0.88 ppm respectively and 87.6 and 142.5ppm against *B. alexandrina* followed by *Ziziphus spina- chriti* which showed molluscicidal activity against *L. cailliaudi* with LC_{50} 311ppm and LC_{90} 500 ppm and caused no mortality of *B. alexandrina* up to1000 ppm. The least active was *Enterolobium contortisiliquum* which gave negative results against both snail species up to1000 ppm. Further purification of active compounds present in *Euphorbia aphylla* and *Ziziphus spina- chriti* may eventually be of great value for the control of snails' intermediate hosts of fascioliasis and schistosomiasis.
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1. Introduction

Schistosomiasis remains as one of the world's most prevalent diseases (King and Dangerfield-Cha, 2008). It is estimated to infect 207 million people worldwide. Approximately tenth of the world population are living with the risk of infection (WHO, 2010). In Egypt, the disease is not only a prime health problem, but it is also an economic one, as it affects million of farmers at the early age diminishing their productivity and exerting a serious socioeconomic problem(El-Baz *et al*.,2003). *Biomophalaria alexandrina* as specific intermediate host of *Schistosoma mansoni* is prevalent in both Upper and Lower Egypt (WHO, 2002).

Fascioliasis is a worldwide zoonotic disease caused by another trematode parasite of the genus *Fasciola* that infects over 17 million people causing significant morbidity and mortality (Mas-Coma *et al*., 2005; WHO, 2006). In Egypt, fascioliasis becomes hyperendemic and problematic where animal reservoir and snail vector are available (Rashed *et al*., 2008). Nearly 24 million Egyptians are at risk and about 800 000 suffering from fasciolosis (WHO, 1995, Haseeb *et al*., 2002).Human infection causes serious hepatic pathological consequences (Soliman, 2008). In addition, fascioliasis is responsible for economic losses estimated at around one billion Egyptian pounds per year (Haseeb *et al*., 2002). In general, El Shazly *et al.* (2002) found concomitant infection between fascioliasis and Schistosomiasis *mansoni*.

Treatment of *Schistosoma* and *Fasciola* 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 drugs with frequent side effects (Ismail *et al.*, 1999 and Abdul-Samie *et al.,* 2010). Therefore snail control is considered not only complementary but essential in *Schistsoma* and *Fasciola* control. It is regarded as a rapid and efficient method for reducing or eliminating transmission and is among the methods of choice to bring these diseases under an adequate control through the breakage of the life cycle of the parasite (Mello-Silva *et al.*, 2006; Jigyasu and Sing, 2010).

Today, mollusciciding is regarded as an important aggressive strategy in the control of the snail hosts of these diseases (Giovanelli *et al*., 2001, Mello-Silva *et al*., 2006). Unlike the use of synthetic drugs, the uses of molluscicides prevent reinfection of people after treatment (WHO, 1993). Copper sulfate and niclosamide were used in Egypt within a program developed by Bayer AG, however, due to their hazardous enviromental effects ,their toxicities to non-target organisms and even man, they were stopped (WHO, 2002 ; Abdelrazek *et al*., 2007).Therefore the search for alternative molluscicides is still ongoing. During recent years much attention has drawn for the use of molluscicides of plant origin. The use of plants with molluscicidal properties appears to be a simple, inexpensive and safe alternative (Singh and Singh, 2010; Al-Daihan, 2010). Also, there is a continuous need to search for new plant species with ideal molluscicidal properties (Tantawy *et al*., 2004; Bakry and Hamdi, 2007). In Egypt, several local plant species screened and proved to have molluscicidal properties against different snail species e.g. *Ambrosia maritime* (Abou Basha ***et a1***., 1994), *Solanum* species (Tantawy *et al*., 2000), *Commiphora molmol* (Abd-Allah *et al*. 2009), *Guayacum officinalis*, *Calatropis procera* and *Euphorbia splendens* (Bakry, 2009).

Euphorbia is the largest genus of flowering plants in the Egyptian flora (El-Karemy, 2008). Over the past twenty years, they have received considerable phytochemical and biological attention (Wu *et al*., 2009). According to Mwine (2011) a good number of *Euphorbia* species are actually potent as medicinal plants and their extracts have been isolated and patented as modern drugs. They have a variety of uses, such as for the treatment of intestinal parasites (Appendino and Szallasi, 1997 and Shi *et al*., 2008). They also possess antiamoebic (Tona *et al*., 2000); anti-plasmodial (Tona *et al*., 2004) and anti-leishmanial activity (Ahmed *et al*., 2006).Earlier studies indicated that the euphorbiales have molluscicidal activity (Tantawy *et al*., 2004; Sermsart *et al*., 2005; Bakry 2009; Singh and Singh, 2010). Alkaloids and saponin are reported among active compounds of several *Euphorbia* species (Siddiqui *et al*., 2009).

Ziziphus spina-christi is one of the plants most commonly used in Egyptian folk for treatment of different diseases and is traditionally used in Arab countries as a medicinal plant (Rigal *et al*., 2006 and Nawash and Al-Horani, 2011). In field of parasitology, the ethanolic extract of *Ziziphus spina-christi* root showed anti-schistosomal activity(Aly *et al*.,2006,El - Rigal *et al*.,2006).Anti-*leishmania* activities of ethanolic and aqueous extracts of the leaves have also been reported (Tonkal *et al*., 2005). The phytochemical composition of *Ziziphus spina- cristi* reported the presence of four saponin glycosides and alkaloids (Shahat *et al*., 2001 and Anthony, 2005).

Enterolobium contortisiliquum is an important species of the family Fabaceae. The essential oil of *Enterolobium contortisiliquum* seeds had been reported to have an antimicrobial activity (Shahat *et al*., 2008). The plant was reported to be rich in saponin (Mimaki *et al*., 2004), a substance responsible for molluscicidal activity (Hostettmann *et al*., 1982, Osman *et al*., 2007).

It is now well established that in many plants the molluscicidal activity is due to the presence of saponin contents (Rawi *et al*., 1996, Osman *et al*., 2007 and Singh and Singh 2010) and alkaloid components (Melendez and Capriles, 2002, El-Ansary *et al*., 2003, Ahmed and Rifaat 2005, Silva *et al*., 2005 and Singh *et al*., 2010). According to these authors, plants containing one or more of these compounds are among the most promising for controlling schistosomiasis and fascioliasis.

Based on these facts and since *Euphorbia aphylla* (Euphorbiaceae), *Ziziphus spina*-christi (Rhamnaceae) and *Enterolobium contortisiliquum* (Fabaceae) have been described as plants rich in saponin and /or alkaloids. The present study is aimed to evaluate the molluscicidal activity of the ethanolic extracts of these plants against *Biomophalaria alexandrina* and *Lymnaea cailliaudi* (*nalatensis*) the snails' intermediate hosts of *Schistosoma mansoni* and *Fasciola* species respectively in a trial to open new areas of application of extracts of these plants as eco-friend molluscicides.

2. Materials and Methods: - Snails:

Laboratory bred uninfected adult *Biomphalaria alexandrina* snails (6-8mm in diameter) and *Lymnaea cailliaudi* (*nalatensis*) (8-10mm in shell length) from the stock reared in Medical Malacology Department, Theodor Bilharz Research Institute (TBRI) were used.

Plant Material:

The plants used in this study were *Euphorbia aphylla* (Family Euphorbiaceae), *Ziziphus spina-christi* (Family Rhamnaceae) and *Enterolobium contortisiliquum* (Family Fabaceae).The plant materials were collected locally from Faculty of Agriculture, Assiut University. Plant species was kindly identified and extracted by Prof. Dr. Zedan Z. Ibraheim, Pharmacognacy Department, Faculty of Pharmacy, Assiut University.Voucher specimens of each plant were kept in the Museum of Pharmacognacy Department, Faculty of Pharmacy.

Preparation of Plant Extracts:

The aerial parts of *Euphorbia aphylla*, Ziziphus *spina- christi* and the mature ripe fruit of *Enterolobium contortisiliquum* were cleaned, cut into small pieces and dried in shade then grounded using blender. About 250 g of air dried powdered plant material was extracted with ethanol (70%), filtered and distilled off under vacuum at temperature not exceeding 50 °C and the residues were stored in dry glass bottles (Bakry, 2009).

Preparation of Molluscicide Solutions:(According to Singab *et al*., 2006)

Stock solutions of 1000 ppm were freshly prepared by dissolving 1 g of each ethanolic extract in the minimal amount of dimethylsulfoxide (DMSO), and made up to 1000 ml by adding dechlorinated water. A series of concentrations (0.25 – 1000 ppm) were prepared from the stock solution of *Euphorbia aphylla* and double serial concentrations (100-200-400 etc.) were prepared from the stock solutions of *Ziziphus spina- christi* and *Enterolobium contortisiliquum*

Determination of molluscicidal activity:

WHO, 1965 guideline was followed for evaluation of the molluscicidal activity of the extracts.A series of exploratory experiments were conducted using the previously prepared concentrations to determine the toxicity range of the plant extracts against the tested snails. Once the extent of the toxicity range was determined, several intermediate concentrations were prepared from the stock solutions (diluted with dechlorinated water) to give mortalities between0-100% according to Osman *et al*., 2007.

For each experimental concentration three replicates were prepared, each of 10 snails/ L. Another three replicates were prepared in dechlorinated water as control. Snails were exposed to the molluscicide suspension for 24 hours at room temperature (exposure period). The tested snails were then left in water for another 24 hour and examined to assess mortality (Recovery period). Snails were considered dead if they probed and remained motionless or if the shell looked discoloured. Mortality rates were recorded. Probit regression analysis (SPSS version 7) aimed to determine the LC_{50} and LC_{90} values as well as their 95 % confidence limits were carried out according to Finney (1971).

3. Results

Molluscicidal activity of *Euphorbia aphylla*:

The effect of various concentrations of ethanol extract of the aerial portion of *Euphorbia aphylla* on adults of *Biomophalaria alexandrina* and *Lymnaea cailliaudi* (*nalatensis*)after 24 hour exposure are listed in tables 1and 2.

The LC_{50} and LC_{90} of this extract against *Biomophalaria alexandrina* after 24 hour exposure were 87.6 and 142.5ppm respectively. While the LC_{50} and LC_{90} of the same extract against *Lymnaea cailliaudi* (*nalatensis*) after 24 hour exposure were 0.66 and 0.88 ppm respectively.

There was a significant difference between molluscicidal activities of ethanol extract of *Euphorbia aphylla* against both snails. *Lymnaea cailliaudi* (*nalatensis*) were more sensitive to *Euphorbia aphylla* extract than *Biomophalaria alexandrina* adults.

The probit mortality showed that the response of the two snail species illustrated a linear relationship with the concentrations (dose ⁄ ppm) of the ethanol extract of *Euphorbia aphylla* as revealed in figure1, 2.The exposed snail species responded differently to different concentrations of the studied plant exract.

Molluscicidal activity of *Ziziphus spina christi*:

Molluscicidal effect of ethanol extract of the aerial portion of *Ziziphus spina- christi* on *B. alexandrina* gave negative results up to 1000ppm.

Molluscicidal effect of ethanol extracts of *Ziziphus spina christi* on *L. cailliaudi* showed LC_{50} 311ppm and LC_{90} 500 ppm. The probit mortality showed that the response of *L. cailliaudi* illustrated a linear relationship with the concentrations (dose ⁄ ppm) of the ethanol extract of *Zizyphus spina-christi* as revealed in figure 3.

Molluscicidal activity of *Enterolobium contortisilicuum*:

Ethanol extract of the fruit of *Enterolobium contortisilicuum* (Family Fabaceae) gave negative results against both snail species up to1000 ppm.

Table (1): Mortality rates, LC_{50} and LC_{90} of ethanolic extract of *Euphorbia aphylla* against *Biomophalaria alexandrina*

Conc. (ppm).	Number of tested snails	Number of dead snails	Mortality rates (%)	LC_{50}	LC_{90}
150	30	30	100	87.6(39.99–332.5)	142.5(98.3– 1092.3)
100	30	12	40		
50	30	9	30		
40	30	6	20		
20	30	0	0		

Probit Transformed Responses

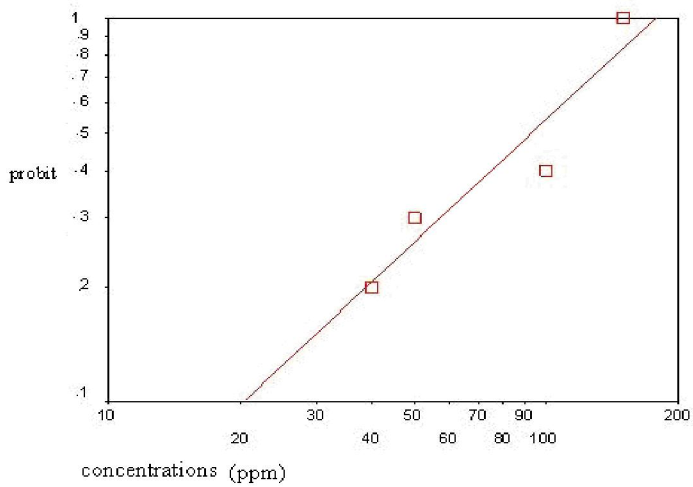

Figure 1.Dose ⁄ probit regression line of *Euphorbia aphylla* on *Biomophalaria alexandrina*

Table (2): Mortality rates, LC_{50} and LC_{90} of ethanolic extract of *Euphorbia aphylla* against *Lymnaea cailliaudi* (*nalatensis*).

Conc.(ppm)	Number of tested snails	Number of dead snails	Mortality rates (%)	LC_{50}	LC_{90}
1	30	30	100	0.66(0.62 - 0.70)	0.88(0.821-0.966)
0.80	30	24	80		
0.75	30	18	60		
0.60	30	12	40		
0.50	30	6	20		
0.25	30	0	0		

Probit Transformed Responses

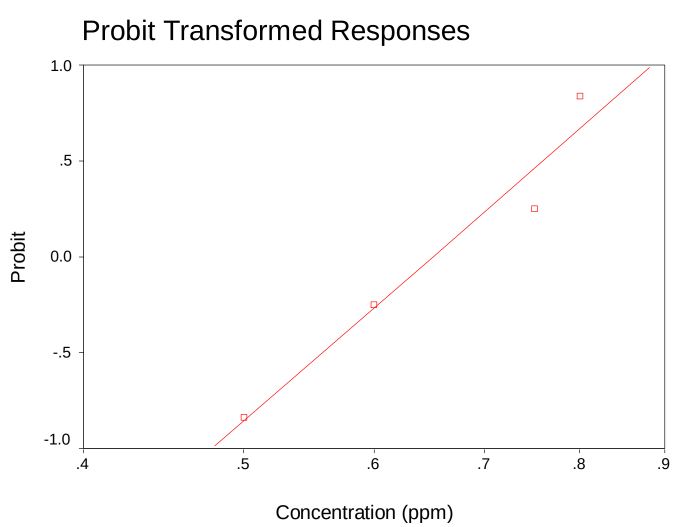

Figure 2. Dose ⁄ probit regression line of *Euphorbia aphylla* on *Lymnaea cailliaudi* (*nalatensis*)

Table (3): Mortality rates, LC_{50} and LC_{90} of ethanolic extract of *Ziziphus spina- christi* against *Lymnaea cailliaudi* (*nalatensis*)

Conc. (ppm)	Number of tested snails	Number of dead snails	Mortality rates (%)	LC_{50}	LC_{90}
800	30	30	100	311(163.83-465.68)	500(384.002 1089.676)
600	30	27	90		
400	30	24	80		
300	30	20	66.66		
200	30	5	16.66		
100	30	0	0		

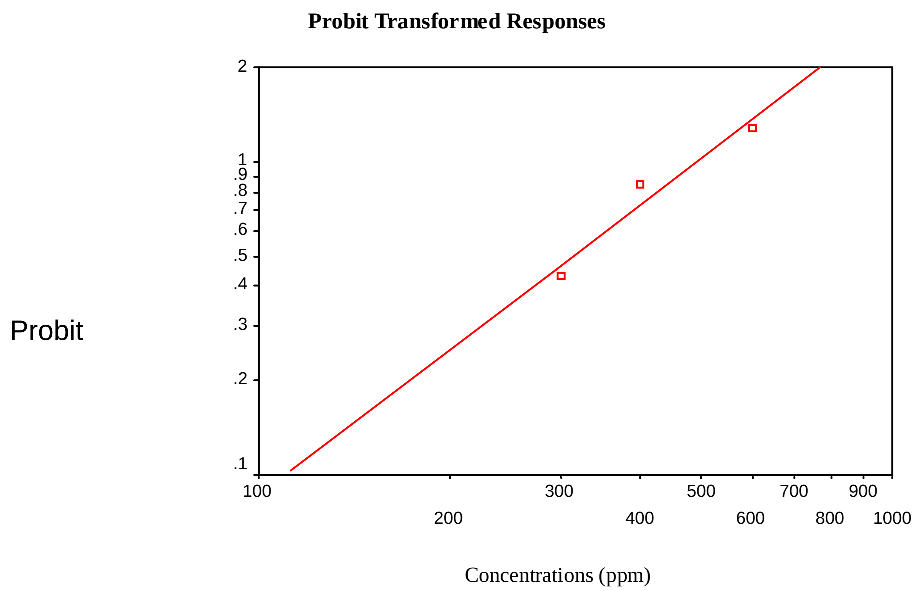

Figure 3.Dose ⁄ probit regression line of *Ziziphus spina-christi* on *Lymnaea cailliaudi* (*nalatensis*).

4. Discussion:

Schistosomiasis and fascioliasis are worldwide parasitic diseases infecting 207 and 17 million people respectively causing significant morbidity and mortality (WHO, 2006, 2010).

In Egypt, positive association between liver fluke infection and schistosomiasis was detected in several governorates. It was explained by the co-existance of both parasites intermediate hosts inhabiting the same type of water bodies(Haseeb *et al*., 2002).Once, these snails intermediate host destroyed, the life cycle will be disrupted (Hamed ,2010; Jigyasu and Sing, 2010). To achieve this goal, different synthetic molluscicidal compounds were used (Essawy *et al*., 2009;Kristoff *et al*., 2010).

The high costs of synthetic molluscicides, their toxicities to non-target organisms and even man as well as the complex organization required in their application, are a major setback to their continued use in schistosomiasis and fascioliasis control programmes. A potential cost effective alternative is the use of compounds from plant origin (WHO, 1993, 2003). Many plants have been screened for their intrinsic molluscicidal properties in an attempt to find an alternative to synthetic ones. Plants containing alkaloids and saponin are among the most promising plants for controlling schistosomiasis (El-Ansary *et al*. 2003, Silva *et al*. 2006; Singh and Singh, 2009; Singh *et al.*, 2010).

Based on these facts and in view of extending problem of schistosomiasis and fascioliasis in terms of morbidity, mortality, treatment cost, it was decided to study three medicinal plant species namely;*Euphorbia aphylla*, *Ziziphus spina-christi*, and *Enterolobium contortisiliquum* for their molluscicidal activity against *Biomophalaria alexandrina* and *L. cailliaudi* (*nalatensis*) according to WHO, 1965 guidelines.

1) Molluscicidal activity of *Euphorbia aphylla*:

Euphorbiaceae is one of the largest families of flowering plants. Members are widely distributed all around the world and some of which are yet to be identified (Sing and Sing, 2010). Molluscicidal activity is widespread in the family Euphorbiaceace, although activity varies greatly from species to species (Al-Zanbagi, 2005, Sharma *et al.,* 2009).

The present study demonstrated that the ethanol extract of *Euphorbia aphylla* possesses molluscicidal activity. These results are in harmony with Mello-Silva *et al*. (2006), Bakry (2009) and Sharma *et al.* (2009) who revealed the molluscicidal activity of different *Euphorbia* species with varying degrees of potency.

In the present study, The LC_{50} and LC_{90} values of *Euphorbia aphylla* against *B. alexandrina* are promising in comparison with some previously studied related plants as *Euphorbia gymnoclada* which did not show a molluscicidal effect against *B. glabrata* (Silva *et al*., 1971). *Euphorbia schimperiana* and *Euphorbia helioscopia* caused no mortality up to 100 ppm. on *Biomphalaria pfeifferi* (Al- Zanbagi 1999). Aqueous extract from *Jatropha curcas* L. (Euphorbiaceae) performed poorly against snails transmitting *Schistosoma mansoni* as 500 ppm caused 50% mortality (Rug and Ruppel, 2000).

Also this activity is better than *Atriplex stylosa, Guayacum officinalis* and *Calatropis procera* with LC_{90s} ranging from 180 to 360 ppm. against Egyptian *B. alexandrina.* On the other hand this activity is lower than that of *E. splendens* (LC_{90} 73 and LC_{50} 40 ppm) (Bakry 2009). These differences in potency can be attributed to several factors including the locality of the plant species, time of collection of the plant sample, part used, storage conditions, method of extraction and solvents type (Brackenbury and Appleton, 1997 and Hassan *et al*., 2010).

The current study was extended to prove the molluscicidal effect of ethanol extract of *Euphorbia aphylla* on *L. cailliaudi* after 24 hours exposure. The LC_{50} and LC_{90} were 0.66 ppm and 0.88ppm respectively. This activity is higher than the latex of *E. hirta* against *Lymnaea acuminate* (LC_{50} 1.29 ppm) (Yadav and Singh, 2011). Also this activity is much higher than that of *Commiphora molmol* oil (LC_{50} and LC_{90} 50 and 85 ppm respectively) (Allam *et al.* 2001), *Phytolacca dodecandra* (Endod) (LC_{90} 2.8 ppm) (Yohannes *et al*., 1979) and *Meryta denhamii* (LC_{50} 26.4 and LC_{90} 70.8 ppm) (Hassan *et al.,* 2010) against *Lymnaea cailliaudi.*

It worth mention that *Commiphora molmol* (Myrrh) is a plant recommended as safe molluscicides(Massoud *et al*.,2004 , Al mathal and Fouad.,2006) and has been licensed for medical use in Egypt and several countries as a fasciolicidal and schistosomicidal drug with high efficacy and safety(Aly and Aly,2006 and Abdul-Samie *et al.,* 2010).Also *Phytolacca dodecandra* is the best studied plant molluscicide (Esser *et al*., 2003).

In the present study, *Lymnaea cailliaudi* has been found to be more susceptible than *Biomophalaria alexandrina* to the toxic action of *Euphorbia aphylla* with the latter requiring high concentrations as lethal doses when compared with the first species. This observation is in accordance with the findings of other investigators using other molluscicides (Allam *et al*., 2001 and Hassan *et al*., 2010). The difference in susceptibility of the two snails to the lethal effect of the same extract could be attributed to the natural resistance of different snail's genera and that the molluscicides may vary in their toxicological effects according to the species of the snails' used (Bakry and Hamdi, 2007 and Osman *et al*., 2007).

Beside its remarkable molluscicidal potency, *Euphorbia aphylla* also presents some very interesting characteristics for an ideal plant molluscicide. It is cosmopolitan and perennial plant. It is not edible to animals and easily cultivable (its multiplication is done by means of asexual reproduction which does not require frequent watering or application of pesticides or fertilizer) (Baptista *et al*., 1997 and Brickell, 2008).

2) Molluscicidal activity of *Ziziphus spina-christi*:

In the present study, the LC_{50} and LC_{90} of ethanol extract of *Ziziphus spina –christi* against *L. cailliaudi* after 24 hours exposure were 311and 500 ppm respectively. This activity is much higher than that reported for the Egyptian plant, *Ambrosia maritima* (damsissa) (LC_{90} 3000 ppm) against Egyptian *Lymnaea cailliaudi* (Abou Basha *et al*., 1994).

In the present study, based on the LC_{50} and LC_{90} values, *Ziziphus spina-christi* demonstrated less potent molluscicidal activity than *Euphorbia aphylla* against *Lymnaea cailliaudi* which can be attributed to the differences in each plant active ingredients, their mode of action and method of penetration of the snails (Rawi *et al*., 1996).

In the present study, *Ziziphus spina –christi* gave negative results up to 1000 ppm on *B. alexandrina*. These results reconfirmed that *Lymnae cailliaudi* is more sensitive. The possibility for the same plant extract to have molluscicidal activity against certain snail species and absence of activity against other species were recorded by Yasuraoka *et al*. (1980) who found that the seeds of *Jatropha carcas* (Family Euphorbiaceae) have a relatively high toxicity against *Oncomelania* while it showed no effect against *Lymnaea* snails.

3) Molluscicidal activity of *Enterolobium contortisiliquum*:

In the present study, *Enterolobium contortisiliquum* is selected due its richness in saponins (Mimaki *et al*., 2004). Saponins have haemolytic properties and toxic effect on most cold-blooded animals including snails and are proved to have molluscicidal activity (Herlt *et al*., 2002, Osman *et al*., 2007 and Singh and Singh, 2009).

In the present study, failure of *Enterolobium contortisiliquum* fruits to produce molluscicidal activity on both snail species up to 1000 ppm. could be attributed to the fact that saponins responsible for its activity are extracted in greater measures with more polar solvents. Supporting this explanation the results obtained by Hassan and Abdel-Rahman (2008) who found that the butanol fraction of *Hedera canariensis* (family Araliaceae) has molluscicidal activity against *Biomophalaria alexandrina* and *Lymnaea cailliaudi.* While ethyl acetate extract of the same plant was inactive. On the contrary, Hassan *et al* (2010) found that the butanol fraction of *Meryta denhamii* flowers which belongs to the same plant family was inactive and ethyl acetate was active against the same snail species.

Conclusion:

The use of *Euphorbia aphylla* may play vital role in controlling schistosomiasis and fascioliasis. The plant is commonly available, easy to collect and prepare for use. Therefore it is the most suitable for biological application which offers a potentially simple, readily available and inexpensive molluscicidal agent of plant origin. In future, more attention should be paid to the mechanism of action of *Euphorbia aphylla* on molluscs and application techniques for its use as plant molluscicides in rural communities. Phytochemical investigations to identify the bioactive ingredient(s) responsible for the molluscicidal potency are recommended. Toxilogical studies on man, fauna and flora of the fresh water are needed to conclude about the possible toxics properties of the ingredient(s).

Results of *Ziziphus spina- christi* suggests further laboratory tests to search for the presence of active component in the different parts o the plant. Such studies would increase their potential for future use as plant molluscicides.

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References:

Abd-Allah K.F., Negm-Eldin M.M., Saleh M.H., El-Hamshary A.M., El-Gozamy B.M. and Aly N.S. (2009): A study on biological control of six freshwater snails of medical and veterinary importance. Egypt. Soc. Parasitol. 39(1):121-39.

Abdel-Rahman E. H. and Hassan S. H. (2008): Molluscicidal saponins from leaves of *Hedera canariensis*. J. Egypt. Soc. Parasitol.38: 293-304.

Abdelrazek1 M. F., Michael1 A. F. and El-Mahrouky F.S. (2007): Synthesis and molluscicidal activity of some new substituted-furan and furo [2, 3- d] pyrimidine derivatives International Journal of Physical Sciences 2 (8): 212-216.

Abdul-Samie R. E., Soliman E. O., El-Nemr H. and Masou A. (2010): Study of Il_1b, Il_4, Il_5 and Ige before and after Mirazid Therapy in children with intestinal schistosomiasis and fascsioliasis. New York Science Journal (3)12 116-122.

Abou Basha L.M., EL Sayad M.H., Allam A.F. and Osman M.M. (1994): The effect of *Ambrosia maritime* (Damsissa) on the viability of *Lymnaea cailliaudi*, an experimental study. J. Egypt. Soc. Parasitol. 24 (3): 513 – 517.

Ahmed A. H. and Rifaat M.M. (2005): Effects of *Solanum nigrum* leaves water extract on the penetration and infectivity of *Schistosoma mansoni* cercariae. J. Egypt. Soc. Parasitol. 35(1): 33–40.

Ahmad I., Farrukh A. and Mohammad O. (2006): Modern Phytomedicine turning medicinal plants into drugs. Wiley- V C H York 136 pp.

Al-Mathal E.M. and Fouad M. (2006): Effect of *Commiphora molmol* on adults, egg masses and egg deposition of *Biomphalaria arabica* under laboratory conditions. J. Egypt. Soc. Parasitol. 36: 305-14.

Al-Daihan S. (2010): Effect of plant molluscicides on selected enzymes related to energy metabolism in *Biomphalaria arabica* snails, molluscan hosts to *Schistosoma mansoni* in Saudi Arabia. J. Egypt. Soc. Parasitol. 40: 187-195.

Allam A. F., El-Sayed M.H and Khalil S. S. (2001): Laboratory assessment of the molluscicidal activity of *Commiphora molmol* (Myrrh) on *Biomphalaria alexandrina*, *Bulinus truncatus* and *Lymnaea cailliaudi.* J. Egypt. Soc. Parasitol. 31(3):683-690.

Aly H.F. and Aly S.A. (2006): Essential role of *Citrus reticulata* and Mirazid in treatment of *Schistosoma mansoni* infected mice: Biochemical and parasitological studies. Polish J. Food Nutr. Sci. 4: 461-467.

Al-Zanbagi N.A. (1999): Plant molluscicides from Saudi Arabian Euphorbiales for use against the snail intermediate hosts of Schistosomes. Ph. D.

Thesis, University of Wales, Aberystwyth.

Al-Zanbagi N. A. (2005): Two molluscicides from Saudi Arabian Euphorbiales against *Bulinus wrighti*. J.K.A.U: Sci., 17:11-19.

Anthony C.C. (2005): Review of *Zizyphus spina-christi* www. Cornelius. Co. uk.

Appendino G. and Szallasi A. (1997): Euphorbium: Modern research on its active principle, resiniferatoxin, revives an ancient medicine. Life Sci. 60, 681–696.

Bakry F.A. (2009): Use of Some Plant Extracts to Control *Biomphalaria alexandrina* snails with emphasis on some biological effects. World Applied Sciences Journal, 3(1): 1335-1345.

Bakry F.A. and Hamdi S.A.H. (2007): Molluscicidal activity of latex aqueous solution of *Euphorbia acetonitril* and *Euphorbia granulate* against the intermediate hosts of Schistosomiasis and Fascioliasis, Journal of Union of Arab Biologists, 27: 101–126.

Baptista D.F, Vasconcellos M.C., Lopes F.H., Paz I.S. and Schall V.T. (1997): Vegetation development and seed germination of *Euphorbia splendens var hislopii* (Euphorbiaceae).A biomolluscicidal species, Arq Biol Tecnol. 40: 435-441.

Barakat R., El-Morshedy H. and Fenwick A. (2005): Efficacy of myrrh in the treatment of human schistosomiasis *mansoni*. Am J Trop Med Hyg .73(2): 365-367.

Brackenbury T. D. and Appleton C. C. (1997): Acute toxicity evaluation of the plant molluscicide, *Apodytes dimidiata*(Icacinaceae), to *Eisenia fetida* (Oligochaeta) and *Oreochromismossambicus* (Cichlidae) in South Africa, Acta Tropica 63 (1):1–14.

Brickell C. and Cathey H. M. (2008): A-Z Encyclopedia of Garden Plants. DK Publishing, Inc. New York, 383.

El-Ansary A., Sammour E.M., Soliman M.S. and Gawish F.A. (2001): In vivo attenuation of *Schistosome* cercarial development and disturbance of egg laying capacity in *B. alexandrina* using sublethal concentrations of plant molluscicides. J. Egypt Soc. Parasitol., 31: 657-669.

El-Baz M. A. Morsy M., EL-Bandary M. and Motaweae S.M. (2003): Clinical and parasitological studies on the efficacy of Mirazid in treatment of *Scistosomiasis haematobium* in Tatoon, Esta center, El-Fayoum Governorate. J.Egypt.Soc.Parasitol. 33:761-776.

El-Karemy Z. (2008): On the taxonomy of the genus *Euphorbia* (Euphorbiaceae) in Egypt. Feddes Repertorium 105 (5-6): 271 – 281.

El-Rigal N. S., Aly S. A , Rizk M. and Said A. (2006): Use of Ailanthus altissima and *Zizyphus spina-christi* extracts as folk medicine for treatment of some hepatic disorders in *Schistosoma mansoni* infected mice. Trends in medical research 1(2):100-112.

El-Shazly A.M., El-Wafa S.A., Haridy F.M., Soliman M., Rifaat M.M. and Morsy T.A. (2002): Fascioliasis among live and slaugthered animals in nine centers of Dakahlia Governorate. J Egypt Soc Parasitol 32(1) 47-57.

Essawy A.E., Abdelmeguied N.E., Radwan M.A., Hamed S.S. and Hegazy A.E. (2009): Neuropathological effect of carbamate molluscicides on the land snail *Eobania vermiculata*. Cell Biol. Toxicol., 25: 275-29.

Esser K.B., Semagn K. and Wolde-Yohannes L. (2003): Medicinal use and social status of the soap berry endod (*Phytolacca dodecandra*) in Ethiopia. J. Ethnopharmacol. 85: 269-277.

Finny D.J. (1971): Probit analysis, third ed., Cambridge University Press, London.

Giovanelli A., Silva C.L., Medeiros L. and Vasconcellos M.C. (2001): The molluscicidal activity of the latex of *Euphorbia splendens* var. *hisloppii* on *Melanoides tuberculata* (Thiaridae), a snail associated with habitats of *Biomphalaria glabrata* (Planorbidae). Mem. Inst. Oswaldo Cruz, 96:123-125.

Hamed M.A. (2010): Strategic control of *Schistosome* intermediate host. Asian J. Epidemiol. 3(3):123-140.

Haseeb A.N., El-Shazly A.M., Arafa M.A. and Morsy A.T. (2002): A review on fascioliasis in Egypt. J. Egypt. Soc. Parasitol. 32(1): 317-354.

Hassan S. E., Abdel-Rahman E. H. and Abdel-Monem A. R. (2010): molluscicidal activity of butanol fraction of *Meryta denhamii* flowers against *Biomophalaria alexandrina* and *Lymnaea nalatensis* .Global Veteriniria 4(1):15-21.

Herlt J.H., Mander L. N. Pongoh E., Rumampuk R.J. and Tarigan P. (2002): Two major saponins from seeds of *Barringtonia asiatica*: Putative antifeedant toward *Epilachna* sp. larvae. J.Nat.Prod. 65: 115-120.

Hostettmann K., Kizu H. and Tomimori T. (1982): Molluscicidal properties of various saponins. Planta Medica, 44: 34-35.

Ismail M., Botros S., Metwally A., William S., Farghally A., Tao L., Day, T.A. and Bennett J. L. (1999): Resistance to praziquantel: direct evidence from *Schistosoma mansoni* isolated from Egyptian villagers. American Journal of Tropical Medicine and Hygiene 60: 932–935.

Jigyasu H. V. and Sing V.K. (2010): Effect of environmental factors on the fecundity, hatchability and survival of snail *Lymnaea*(Radix) *acuminate*(Lamarck):vector of fascioliasis. J. Water and Health, 8:109-115.

King C .H .and Dangerfield-Cha M.(2008): The unacknowledged impact of chronic schistosomiasis, Chronic Illness 4:65–79.

Kristoff G., Guerrero N.R.V. and Cochon A.C. (2010): Inhibition of cholinesterases and carboxylesterases of two invertebrate species, *Biomphalaria glabrata* and *Lumbriculus variegates* by the carbamate pesticide carbaryl. Aquat. Toxicol. , 96: 115-123.

Mantawy M.M., Hamed M.A., Sammour E.M and Sanad M. (2004): Influence of *Capparis spinosa* and *Acacia arabica* on certain biochemical haemolymph parameters of *Biomphalaria alexandrina*. J. Egypt. Soc. Parasitol., 34: 659-677.

Mas-Coma S., Bargues M.D., Valero M.A. (2005): Fascioliasis and other plant-borne trematode zoonoses. Int J Parasitol 35: 1255-1278.

Massoud A. M., El-Kholy N.M. El-Shennawy F.A. and Farag R.E. (2004): Study of some immune aspects in patients with fascioliasis before and after *Commiphora molmol* (Mirazid) treatment. J. Egypt. Soc. Parasitol., 34:315-332.

Melendez P.A. and Capriles V.A.(2002): Molluscicidal activity of plants from Puerto Rico.Ann Trop.Med parasitol 96(2):209-18.

Mello-Silva C. C.,Vasconcellos M. C., Pinheiro, J. and Rodrigus M. L.A. (2006): Physiological changes in *Biomphalaria glabrata* (Say), (Pulonata: Planorbidae) caused by sublethal concentrations of the latex of *Euphorbia splendens var. hisloppii* N. E.B. (Euphorbiaceae). Mem Inst Oswaldo Cruz. 101 (1): 3-8.

Mimaki Y., Harada H., Sakuma C., Haraguchi M., Yui S. , Kudo T. ,Yamazaki M. and Sashida Y.(2004): Contortisiliosides A-G: Isolation of seven new triterpene bisdesmosides from *Enterolobium contortisiliquum* and their cytotoxic activity .Helvetica Chimica Acta .87 (4): 851 - 865

Mwine J. T. (2011): Evaluation of pesticidal properties of *Euphorbia tirucalli* L. (Euphorbiaceae) against selected pests. Ph. D. Thesis, Faculty of Bioscience Engineering, Ghent University, Belgium.

Nawash S.O. and Al-Horani S.A. (2011): The most important medicinal plants in Wadi Araba desert in South West Jordan: A review article. Advances in Environmental Biology, 5(2): 418-425.

Nihei K., Ying B.P., Murakami T., Matsuda H., Hashimoto M. and Kulso I. (2005): Pachvelasides and novel molluscicidal triterpene saponins from *Pachyelasma tessmanni*. J. Agric. Food Chem., 53: 608-613.

Osman E.A., Mohamed E.M., Abu Elreesh B.I. and Elegami A.A. (2007): Molluscicidal activity of *combretum glutinosum*. International journal of molecular medicine and advance sciences 3 (4): 151-154.

Rashed A. A., Khalil H.M. H. and Morsy A. T.A. (2008): Zoonotic ectopic Fascioliasis: review and discussion J. Egypt. Soc. Parasitol., 38 (1):591 – 608.

Rawi S.M., El-Gindy H. and Abdel Kader A. (1996): New possible molluscicides from *calendula Micrantha officinalis* and *Ammi majus*. II. Molluscicidal, physiological and egg laying effects against *Biomphalaria alexandrina* and *Bulinus truncates*. Journal of Ecotoxology and Environmental Safety 35:261-267.

Rawi S.M., Al-Hazmi M. and Al-Nassr F.S. (2011): Comparative study of the molluscicidal activity of some plant extracts on the snail vector of *Schistosoma mansoni*, *Biomphalaria alexandrina*. Int. J. Zool. Res., 7: 169-189.

Rug M. and Ruppel A. (2000): Toxic activities of the plant *Jatropha curcas* against intermediate snail hosts and larvae of schistosomes. Trop. Med. and International Health. 5(6): 423-430.

Sermsart B., Sripochang, S., Suvajeejarun, T., Kiatfuengfoo, R. (2005): The Molluscicidal activity of some *Euphorbia milli* hybrids against the snail *Indoplanorbis exustus*. Southeast Asian J. Trop. Med. pub .health 36(4):192-5.

Shahat A. A., El-Barouty G., Hassan R. A., Hammouda F .M., Abdel-Rahman F.H.and Mahmoud A. S.(2008):Chemical composition and antimicrobial activities of the essential oil from the seeds of *Enterolobium contortisiliquum* Journal of Environmental Science and Health, Part B, 43(6):519 – 525.

Shahat A.A. , Pieters L., Apers S. , Nazeif N.M., Abdel-Azim N. S., Berghe D.V. and Vlietinck A.J.(2001):Chemical and biological investigations on *Zizyphus spina-christi* L. Phytother. Res. 15, 593–597

Sharma S., Singh T. and Vijayvergia R. (2009): Molluscicidal activity of some medicinal plants. Journal of Herbal Medicine and Toxicology 3 (2) 155-157.

Shi Q.W., Su X.H. and Kiyota H. (2008): Chemical and pharmacological research of the plants in genus *Euphorbia*. Chem. Rev., 108, 4295–4327.

Siddiqui S., Verma A., Rather A. A., Japeen F. and Meghvansi M.K. (2009): Preliminary phytochemicals analysis of some important medicinal and aromatic plants.Advances in biological research 3(5-6):188-195.

Silva M .J.M., Souza M.P. and Rouquayrol M.Z. (1971): Atividade moluscicida de plantas do Nordeste brasileiro(II) Revista Brasileira de Farmacia,52: 117-123.

Silva T.M, Camara C.A., Agra M .F, De Carvalho M.G., Frana M.T., Brandoline S.V., da Silva Paschoal L. and Braz-Filho R.

(2005):Molluscicidal activity of *Solanum* species of the Northeast of Brazil on *Biomphalaria glabrata*. Fitoterapia 77: 449-452.
Singh A. and Singh V.K. (2009): Molluscicidal activity of *Saraca asoca* and *Thuja orientalis* against the fresh water snail *Lymnaea acuminata*. Vet. Parasitol. 164: 206-210.
Sing S.K. and Sing A. (2010): Metabolic changes in freshwater harmful snail *Lymnaea acuminata* due to aqueous extract of bark and leaf of *Euphorbia pulcherima* plant. Am. Euras. J. Toxicol. Sci., 2 (1): 13-19.
Singh S .K. , Yadav R.P. and Singh A. (2010): Molluscicides from some common medicinal plants of Eastern Uttar Pradesh, India. J. Applied Toxicol. 30: 1-7.
Singab A. N. B., Ahmed A. H. Sinkkonenc J., Ovcharenkoc V. and Pihlajac K. (2006): Molluscicidal activity and new flavonoids from *Iris germanica* L. Z. Naturforsch. 61(c): 57 - 63.
Soliman F.M. (2008): Epidemiological review of human and animal fascioliasis in Egypt. J Infect Developing Countries 2(3):182-189.
Tantawy A., Sharaf El-Din A.T., Bakry F.A.(2000):Laboratory evaluation of the mollusciciding activity of *Solanum dubium* (Solanaceae) against *Biomphalaria alexandrina* snails, The Journal Proceeding of the First International Conference on Biological Science Egypt 1 307–318.
Tantawy A., Mostafa B.B. and Sharaf El-Din A.T.(2004):Molluscicidal activity of *Synadenium grantii* (Euphorbiaceae) against *Biomphalaria alexandrina* and *Bulinus truncatus* the intermediate host snails of schistosomiasis in Egypt and their infectivity with the parasite, Egyptian Journal of Science 14 183–196.
Tona L., Kambu K., Ngimbi N., Mesia K., Penge O., Lusakibanza M., Cimanga R.K., De Bruyne T., Apers S., Totte J., Pieters L. and Vlietinck A. J. (2000): Antiamoebic and spasmolytic activities of extracts from some antidiarrhoeal traditional preparations used in Kinshasa, Congo. Phytomedicine. 7(1): 31-38.
Tona L., Cimanga R.K, Mesia K., Musuamba C.T., Bruyne T.D., Apers S., Hernans N., Van Miert S., Pieters L., Totte J.and Vlietinck A.J. (2004): In vitro antiplasmodial activity of extracts and fractions from seven medicinal plants used in the Democratic Republic of Congo. J. Ethnopharmacol. 93: 27-32.
Tonkal A., Soliman S.H. Jamjoom B.M., Altaieb M. A., and Al-Bar A.H. (2005): Preliminary study on the effect of *Ziziphus spina-christi* on selected *Leishmania* species Sc. J. Az. Med. Fac. (Girls) 26(1):1915- 1921.
WHO (1965): Molluscicide screening and evaluation, Bulletin of the World Heath Organization 33: 567–581.
WHO (1993): The control of Schistosomiasis, Tech. Rep. 830, WHO, Geneva, Switzerland.
WHO (1995): Control of foodborne trematode infections, Technical Sheet No. 849, Geneva.
WHO (2002): Niclosamide Technical Material, WHO Specifications, p.599
WHO (2003): Action against worms, report series, ISSUE1.
WHO (2006): Report of the WHO informal meeting on use of triclabendazole in fascioliasis control, WHO headquarter, Geneva, Switzerland.17-18 October.
WHO (2010): Schistosomiasis, Fact Sheet No. 115, February http: // www. WHO. int / mediacentre/factsheets/fs115/en.
Wu C. Q ., Tang P. Y. , Ding W. A., You Q.F., Zhang L. and Duan A. J. (2009): C-NMR Data of three important Diterpenes isolated from *Euphorbia* species. Molecules, 14: 4454-4475.
Yadav P. R. and Singh A. (2011): Efficacy of *Euphorbia hirta* latex as plant derieved molluscicide against freshwater snails Rev. Inst. Med. Trop. Sao Paulo53 (2):101-106.
Yasuraoka K., Hashiguchi J. and Bias B.L.(1980): Laboratory assessment of the molluscicidal activity of the plant *Jatropha Carcas* against *Oncomelania* snails, proceedings of the Philippine-Japan joint Conference on Schistosomiasis Research and Control, Manila, The Japan International Cooperation Agency, 110-112.
Yohannes W., Lemma T. and Lemma A., (1979): New approaches to Ended (*Phytolacca dodecandra*) extraction, SIIVET: Ethiopian J. Sci. 2(2):121-127.

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