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Biological and biochemical responses of infected *Biomphalaria alexandrina* snails with *Schistosoma mansoni* post exposure to the pesticides Basudin and Selecron and the phytoalkaloid Colchicine

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This study aims to detect the molluscicidal properties of the pesticides Basudin and Selecron and the phytoalkaloid Colchicine against *Biomphalaria alexandrina* snails including their infection with *Schistosoma mansoni*, production of cercariae, the levels of total protein, globulin and albumin and the activities of alanine and aspartate transaminases (AIT and AsT) and acid and alkaline phosphatases (AcP and AkP) enzymes in tissues of treated snails. The molluscicide Bayluscide was used as a reference compound. After 24 h of snails exposure to the tested compounds, Selecron was the most toxic one. Moreover, about 96% death rates of *S. mansoni* miracidia and cercariae were recorded after 45 min of exposure to the snails' LC_{90s} of Basudin and Selecron. In addition, infection rates of snails with *S. mansoni* and cercarial production were reduced post their exposure to LC₁₀ and LC₂₅ of Basudin, Selecron and Bayluscide and to 100 and 250 ppm Colchicine either during or post snails exposure to miracidia. Thus, snails exposure to LC₂₅ of Selecron reduced cercarial production from 795.2 cercariae/control snail to 72.5 cercariae/infected treated snail. The results, also, revealed that total protein, globulin and albumin concentrations of treated snails were less than control group, while activities of the enzymes AsT, AIT and AkP were elevated. It is concluded that the tested compounds have deleterious effects on viability of *S. mansoni* miracidia and cercariae, the snails' biochemical parameters, their infection with this parasite and production of cercariae from infected snails. Then, accidental introduction of such chemicals to irrigation system during agricultural activities could negatively interrupt and/or prevent schistosomiasis transmission.

Key words: Pesticides, snails, *Schistosoma mansoni*, transaminases, phosphatases activities.

INTRODUCTION

Schistosomiasis remains one of the most public health problems in many developing countries (Kibiki et al., 2004). The plant pesticides are widely used in agricultural activities (Kreuger, 1999). They usually reach the irrigation and drainage systems causing great deteriorations in the abiotic and biotic components of

such water courses including the snail intermediate hosts of schistosomiasis. Basudin and Selecron are among the currently used organophosphorus pesticides in agricultural activities. They inhibit the activity of acetylcholinesterase which is important for nervous system functions (Van Cong et al., 2006). In rats, Diazinon (Basudin) intraperitoneal administration damaged their testes germinal epithelium leading to spermatogenesis failure after 36 hours, then infertility can appear (Cabaj et al., 2010). Selecron is moderately toxic

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to birds and has low mammalian toxicity with good insecticidal properties (Tomlin, 1997). Colchicine has long been used experimentally to visualize metaphase chromosomes in cytogenetic studies for improvement of plants' and animals' productivity (Molad, 2002).

The changes in total protein, albumin and amino acid metabolism in mice and rats reflect hepatocellular injury induced by the insecticide Profenofos (Gomes et al., 1999; Yousef et al., 2006). The levels of AIT, AsT, AkP, uric acid, creatinine, and blood glucose in the serum of rabbits orally treated with Basudin were significantly increased, whereas total protein and albumin contents were decreased (Salih, 2010).

The molluscicide Bayluscide is more toxic to the free living larval stages of schistosomes than the snail intermediate hosts (Tchoun Wou et al., 1991). Exposure of *Biomphalaria alexandrina* snails to certain chemicals deteriorated the host-parasite relationships, which are highly specific and dependable on their biochemical and physiological activities that could interrupt the transmission of this parasite (El-Ansary et al., 2001).

Therefore, the present study aims to determine the effect of the pesticides Basudin and Selecron and the phytoalkaloid Colchicine against *B. alexandrina* snails, their infection with *Schistosoma mansoni* and cercarial production from infected snails. In addition, the larvicidal potency of these compounds on *S. mansoni* miracidia and cercariae and on certain biochemical parameters of snails' tissues were evaluated.

MATERIALS AND METHODS

Snails

B. alexandrina snails (non infected) were reared in dechlorinated water (25°C) at Medical Malacology Laboratory, Theodor Bilharz Research Institute (TBRI), Imbaba, Giza, Egypt (Liang et al., 1987).

The tested compounds

The pesticides Basudin and Selecron (organophosphorus) were from the Plants Protection Center, Ministry of Agriculture, Dokki, Giza, Egypt, and the phytoalkaloid Colchicine was purchased from El Nasr Pharmaceutical Chemistry Company, Cairo, Egypt. Bayluscide was from Snails Control Division, Ministry of Health, Egypt, as a reference molluscicide.

Schistosoma mansoni cercariae and ova

They were from Schistosome Biological Supply Center (SBSC) at Theodor Bilharz Research Institute (TBRI).

Bioassay tests

Molluscicidal activity

A stock solution of 1000 ppm was prepared from each pesticide

and Colchicine on the basis of w/v using dechlorinated water (pH 7.0-7.5). A series of concentrations was prepared from each compound (WHO, 1965). Three replicates were used, each of ten snails (6-8 mm)/L, for each concentration. Exposure and recovery periods were 24 h each; at 25±1°C. For each test, 3 replicates of control snails were maintained under the same experimental conditions in dechlorinated water. The effectiveness of each compound has been expressed as LC₅₀ and LC₉₀ (Litchfield and Wilcoxon, 1949).

Cercaricidal and miracidicidal activities

To test the cercaricidal effect, five ml of water containing about 100 fresh shedding *S. mansoni* cercariae were mixed with 5 ml of double concentration from each of the tested compounds in a divided Petri dish. The tested concentrations were the snails' LC₀, LC₁₀, LC₂₅, LC₅₀ and LC₉₀ from each pesticide and 100 and 250 ppm of Colchicine. Another 10 ml dechlorinated water containing about 100 fresh shedding cercariae was kept as control. After 15, 30, 45, 60 and 120 min, the cercariae were observed under a dissecting microscope for alterations in their mobility. Stationary cercariae were considered dead and their mortality in the test and control aquaria was recorded (Youssef, 2010). For miracidia, *S. mansoni* ova were allowed to hatch in dechlorinated tap water. Five milliliter of water containing about 100 fresh hatching miracidia was mixed with 5 ml of double concentration as in cercaricidal test. Stationary miracidia were considered dead and the mortality rates were recorded.

Effect on infection of *B. alexandrina* snails with *S. mansoni*

The snails (6-8 mm) were exposed for 24 h to LC₀, LC₁₀ and LC₂₅ of the tested pesticides and to 100 and 250 ppm of Colchicine during miracidial exposure and at 3, 7 and 21 days post-miracidial exposure.

Snails were exposed to miracidia in mass for 24 h under illumination, 10 fresh hatched miracidia/snail. For each concentration three replicates, each of 10 snails/L in glass container, were prepared. After that, the snails were transferred to clean dechlorinated water (25±1°C) and daily fed oven dried lettuce leaves throughout the prepatent and patent periods (Massoud et al., 1973). A control group of three replicates, each of 10 snails/L was exposed to miracidia concurrently with the experimental snails and treated similarly till cercarial emergence. Dead snails were removed daily and surviving snails were individually examined once weekly for cercarial shedding 24 days post-miracidial exposure. Then, cercarial production/ infected snail was recorded (Youssef, 2010).

Effect of biochemical parameters on snails' tissues

B. alexandrina snails (8-10 mm) were exposed for 24 h/week for 4 successive weeks to LC₀, LC₁₀ and LC₂₅ of each pesticide and to 100 and 250 ppm of Colchicine. For each concentration, 30 snails were used and another group was maintained in dechlorinated water (25±1°C) as control. The soft parts of snails were dissected out from their shells after gently crushing, weighed and homogenized in cold distilled water at a ratio of 1 g: 10 ml using a glass homogenizer for 5 min.

The homogenates were centrifuged for 15 min at 3000 rpm (4°C) and the fresh supernatant was then used in enzymes and total protein assays (El-khayat and Abu-Zikri, 2004). The tested biochemical parameters were total protein (Domas, 1975) and albumin (Gustafsson, 1976) concentrations and the activities of

Table 1. Molluscicidal activity of the tested compounds against adult *Biomphalaria alexandrina* snails (24 h exposure).

Compound	LC ₀ ppm	LC ₁₀ ppm	LC ₂₅ ppm	LC ₅₀ ppm	LC ₉₀ ppm	Slope
Basudin	1.41	10.53	12.25	14.16	17.78	1.22
Selecron	0.402	2.44	3.11	4.02	5.57	1.36
Bayluscide	0.02	0.07	0.16	0.25	0.43	1.86
Colchicine	-	-	-	> 500	-	-

transaminases AsT and AIT (Reitman and Frankel, 1957; White et al., 1970) and the phosphatases AcP (Moss, 1984) and AkP (Babson, 1965) enzymes. For evaluating these parameters spectrophotometrically kits from Quimica Clinica Aplicada S. A. (QCA) Ltd., Spain were used.

Statistical analysis

Snails mortality and infection rates were analyzed by Chi-square values of contingency tables (Southwood, 1978). The mean values of prepatent and patent periods, cercarial production/snails and biochemical parameters of the tested and control snails were compared by student "t" test (Goldstein, 1964). Statistical analysis was performed by the SPSS computer program (version 13.0 windows).

RESULTS

From the bioassay test, the pesticide Selecron showed a marked toxic effect against *B. alexandrina* snails compared to Basudin and Colchicine. Its LC₉₀ was 5.57 ppm after 24 h of exposure at 25±1 °C (Table 1).

Effect of the tested compounds on the free living larval stages of *S. mansoni* (miracidia and cercariae)

Table 2 indicated that miracidicidal and cercaricidal potencies of the tested compounds are time and concentration dependent. For miracidia, the mortality rate at LC₅₀ of Selecron was increased from 7 to 57% by elongation of the exposure period from 15 to 60 min. Moreover, 90% death of miracidia was observed after 45 min of exposure to LC₉₀ of Basudin, Selecron and Bayluscide, while no death was seen among specimens of control group. For 250 ppm Colchicine, approximately no toxic effect was noticed after 60 min of exposure (only 9% death).

Concerning cercariae, the data revealed them to be more susceptible to the tested pesticides than miracidia. Thus, more than 90% death of cercariae was noticed post 30 min of exposure to LC₉₀ of each of these compounds, while they exhibited high tolerance to 250 ppm of Colchicine after 60 min.

Effect on infection of *B. alexandrina* snails with *S. mansoni*

Survival rate of the snails at first shedding

From Table 3, 24 h of snails exposure to LC₀, LC₁₀ and LC₂₅ of Basudin, Selecron and Bayluscide and to 100 and 250 ppm Colchicine during and post exposure to *S. mansoni* miracidia significantly reduced their survival rates at 1st shedding and their infection rates compared to their corresponding control groups. Thus snails survival rates post exposure to LC₂₅ of Basudin during- and after 21 days of miracidial exposure were 15 and 20%, respectively, compared to 87.5% for control group (P<0.001). As well, the infection rates at LC₂₅ of Selecron during- and after 3 days of miracidial exposure were 20 and 31.3% respectively, in comparison with 94.4% for control group (P<0.001).

Moreover, raising the concentration of the tested pesticides caused more decrement in survival rates at 1st shedding and infection rates of snails treated either during- or post-miracidial exposure.

Thus, the infection rate of snails exposed to Selecron after 3 days of miracidial exposure was reduced from 66.7 to 31.3% by raising the concentration from LC₁₀ to LC₂₅.

Prepatent period and duration of cercarial shedding

The results showed, generally, an elongation of prepatent period for snails treated with the tested compounds after 7 and 21 days of miracidial exposure. The longest period was seen for snails treated with LC₂₅ of Bayluscide (Figure 1) after 21 days of miracidial exposure, being 62 days compared to 47.5 days for control group (P<0.001).

On the other hand, the duration of cercarial shedding from snails treated with the tested compounds was shorter than their corresponding control groups. The shortest period was recorded for snails treated with Colchicine 250 ppm during-miracidial exposure and LC₂₅ of Basudin after 7 days of miracidial exposure, being 4.5 and 5 days, respectively compared to 25.1 days for control group (P<0.001).

Table 2. Effect of the compounds Basudin, Selecron, Bayluscide and Colchicine on *Schistosoma mansoni* miracidia and cercariae.

Concentration (ppm)	% cumulative mortality of									
	Miracidia (min)					Cercariae (min)				
	15	30	45	60	120	15	30	45	60	120
Control	0	0	0	0	0	0	0	0	1	7
Basudin	LC ₀	0	0	0	1	0	0	3	18	23
	LC ₁₀	0	0	3	5	0	0	13	25	37
	LC ₂₅	0	0	9	23	0	2	19	37	53
	LC ₅₀	0	9	13	49	14	34	61	78	89
	LC ₉₀	60	83	96	100	64	93	96	100	
Selecron	LC ₀	0	0	0	3	0	0	7	13	19
	LC ₁₀	0	0	0	10	0	0	13	20	43
	LC ₂₅	0	0	3	15	0	3	14	25	47
	LC ₅₀	7	19	49	57	12	29	59	67	73
	LC ₉₀	75	95	97	100	75	94	96	100	
Bayluscide	LC ₀	0	0	0	0	0	0	8	9	14
	LC ₁₀	0	0	2	5	0	3	12	15	41
	LC ₂₅	0	0	13	27	0	7	23	37	53
	LC ₅₀	2	8	48	59	0	14	58	69	83
	LC ₉₀	18	87	93	98	10	97	100		
Colchicine	100	0	0	0	4	43	0	0	4	21
	250	0	0	2	9	65	0	0	9	29

Basudin: LC₀ (1.41 ppm), LC₁₀ (10.53) and LC₂₅ (12.25) - Selecron: LC₀ (0.402), LC₁₀(2.44) and LC₂₅ (3.11), Bayluscide: LC₀ (0.02), LC₁₀ (0.07) and LC₂₅ (0.16).

Cercarial production

Table 3 showed that this parameter was reduced by snails treatment with the tested compounds during- and post-miracidial exposure. Thus, the cercarial production/infected snail from groups treated with LC₀ of Basudin and Selecron after 3 days of miracidial exposure was 427.3 and 310.2 cercariae/snail respectively, compared to 887.3 and 811.3 cercariae/snail of their corresponding control groups (P<0.001). The reduction rates under these conditions were 51.8 and 61.8%, respectively. Moreover, this parameter for snails treated with 100 ppm of Colchicine after 3 days of miracidial exposure suffered from more reduction as it scored 227.5 cercariae/snail compared to 887.3 cercariae/control snail (P<0.001), with a reduction rate of 74.4%.

The results, also, revealed more reduction in this parameter for snails treated with the tested compounds during-miracidial exposure compared to those treated post-miracidial exposure. Thus, cercarial production/snail from groups treated with LC₁₀ of Basudin and Selecron during-miracidial exposure was 213 and 62 cercariae/snail respectively, compared to 622.7 and 372.1 cercariae/snail for groups treated after 7 days of

miracidial exposure.

Effect on biochemical parameters in tissues of *B. alexandrina* snails

Determination of biochemical parameters in tissues of *B. alexandrina* snails treated with LC₁₀ and LC₂₅ of Basudin, Selecron and Bayluscide and 100 and 250 ppm of Colchicine indicated a marked decrement in the total protein, globulin and albumin concentrations in comparison with control group (Figure 2). Thus, the total protein concentration for snails exposed to LC₂₅ of Basudin, Selecron and Bayluscide and 250 ppm of Colchicine were reduced to 34.6, 28.6, 35.8 and 36.2 mg/g tissue, respectively, compared to 48.4 mg/g tissue for control group (P<0.01).

However, snails treatment with Basudin, Selecron, Colchicine and Bayluscide raised the activities of the enzymes AsT and AIT in their tissues, but vice-versa was recorded for AcP in comparison with control groups (Table 4). For AkP, its activity was elevated by the tested

Table 3. Effect of the compounds Basudin, Selecron, Bayluscide and Colchicine on survival rate at 1st shedding, infection rate and cercarial production of *Biomphalaria alexandrina* exposed to *Schistosoma mansoni*.

Compound	Concentration (ppm)	During-miracidial exposure			Post-miracidial exposure								
		%	%	No. of cercariae/ snail (a)	3 days			7 days			21 days		
					Survival	Infection	No. of cercariae/ snail (a)	Survival	Infection	No. of cercariae/ snail (a)	Survival	Infection	No. of cercariae/ snail (a)
Basudin	Cnt.	87.5	91.4	887.3±18.61	87.5	91.4	887.3±18.61	87.5	91.4	887.3±18.61	87.5	91.4	887.3±18.61
	LC ₀	55.0***	40.9***	340.0±76.90***	55.0***	45.5***	427.3±96.04***	80.0	75**	403.9±95.18***	55.0***	77.3*	426.7±86.83***
	LC ₁₀	45.0***	38.9***	213.0±55.94***	45.0***	61.1***	758.2±229.9	60.0***	45.8***	256.2±63.95***	45.0***	44.4***	622.7±207.15
	LC ₂₅	15.0***	33.3***	136.5±90.58***	35.0***	35.7***	517.5±172.96*	45.0***	38.9***	694.6±214.27	20.0***	0	-
Selecron	Cnt.	90.0	94.4	795.2±28.25	90.0	94.4	795.2±28.25	90.0	94.4	795.2±28.25	90.0	94.4	795.2±28.25
	LC ₀	65.0***	30.8***	268.8±68.24***	67.5***	63.0***	491.2±110.26*	82.5	51.5***	465.4±53.84***	70.0***	71.4***	439.6±138.82*
	LC ₁₀	30.0***	50.0***	62.0±5.39***	52.5***	66.7***	447.0±149.91*	62.5***	50.0***	486.3±71.16***	55.0***	63.6***	372.1±82.80***
	LC ₂₅	25.0***	20.0***	72.5±17.69***	40.0***	31.3***	301.3±151.47**	45.0***	27.8***	1056.8±391.22	37.5***	33.3***	263.2±118.08***
Bayluscide	Cnt.	82.3	90.9	811.3±37.14	82.3	90.9	811.3±37.14	82.3	90.9	811.3±37.14	82.3	90.9	811.3±37.14
	LC ₀	45.0***	44.4***	274.5±63.22***	65.0**	50***	310.2±66.84***	80.0	62.5***	412.5±68.29***	62.5**	80	386.0±84.05***
	LC ₁₀	45.0***	22.2***	164±55.45***	47.5***	52.6***	766.7±257.50	52.5***	61.9***	743.9±163.25	57.5***	73.9**	678.1±80.67
	LC ₂₅	35.0***	14.3***	302±109.96***	20.0***	0	-	45.0***	27.8***	1302.6±265.38	37.5***	66.7***	535.7±131.08*
Colchicine	Cnt.	87.5	91.4	887.3±18.61	87.5	91.4	887.3±18.61	87.5	91.4	887.3±18.61	87.5	91.4	887.3±18.61
	100 ppm	70.0**	42.9***	170.3±39.20***	60.0***	33.3***	227.5±70.11***	52.5***	52.4***	481.3±73.18***	40.0***	50.0***	330.8±141.77***
	250 ppm	55.0***	54.5***	146.1±31.12***	50.0***	30.0***	175.3±31.12***	47.5***	57.9***	477.9±168.88*	30.0***	50.0***	400.5±103.65***

Basudin: LC₀ (1.41 ppm), LC₁₀ (10.53) and LC₂₅ (12.25) - Selecron: LC₀ (0.402), LC₁₀(2.44) and LC₂₅ (3.11) , Bayluscide: LC₀ (0.02), LC₁₀ (0.07) and LC₂₅ (0.16) *, **& *** = Significantly different from control at p< 0.05, p<0.01 and p<0.001, respectively, (a): Mean± S.E.

pesticides, but decreased by Colchicine treatments.

DISCUSSION

In this study, Selecron was more toxic to the snails than Basudin. Similarly, Youssef (2010) recorded such observation on Selecron and Basudin against *B. alexandrina* snails. The present mortality rates of *S. mansoni* miracidia and cercariae were concentration

and exposure period dependent. This agrees with Mahmoud (2006 a) on the pesticides Regent and Mimic. Similar conclusion was recorded by Hasheesh and Mohamed (2011) against *S. haematobium* miracidia and cercariae exposed to the pesticides Chlorpyrifos and Profenofos.

The present results showed that survival rates of snails at 1st shedding post 24 h of exposure to the tested compounds were significantly less than their corresponding control groups. Similar conclusion was recorded by Massoud et al. (1973) on *B.*

truncatus snails exposed to Bayluscide and infected with *S. haematobium*. This was also, recorded for *B. alexandrina* snails post their subjection to the fungicide Topas (Esmaeil, 2009) and the pesticides Match and Vertimec (Youssef, 2010) after 3 weeks of snails exposure to *S. mansoni* miracidia. However, Hira and Webbe (1972) found that *B. glabrata* snails treated with triphenyl lead acetate after 20 days of exposure to *S. mansoni* miracidia, had a similar mortality rate at 1st shedding as control group.

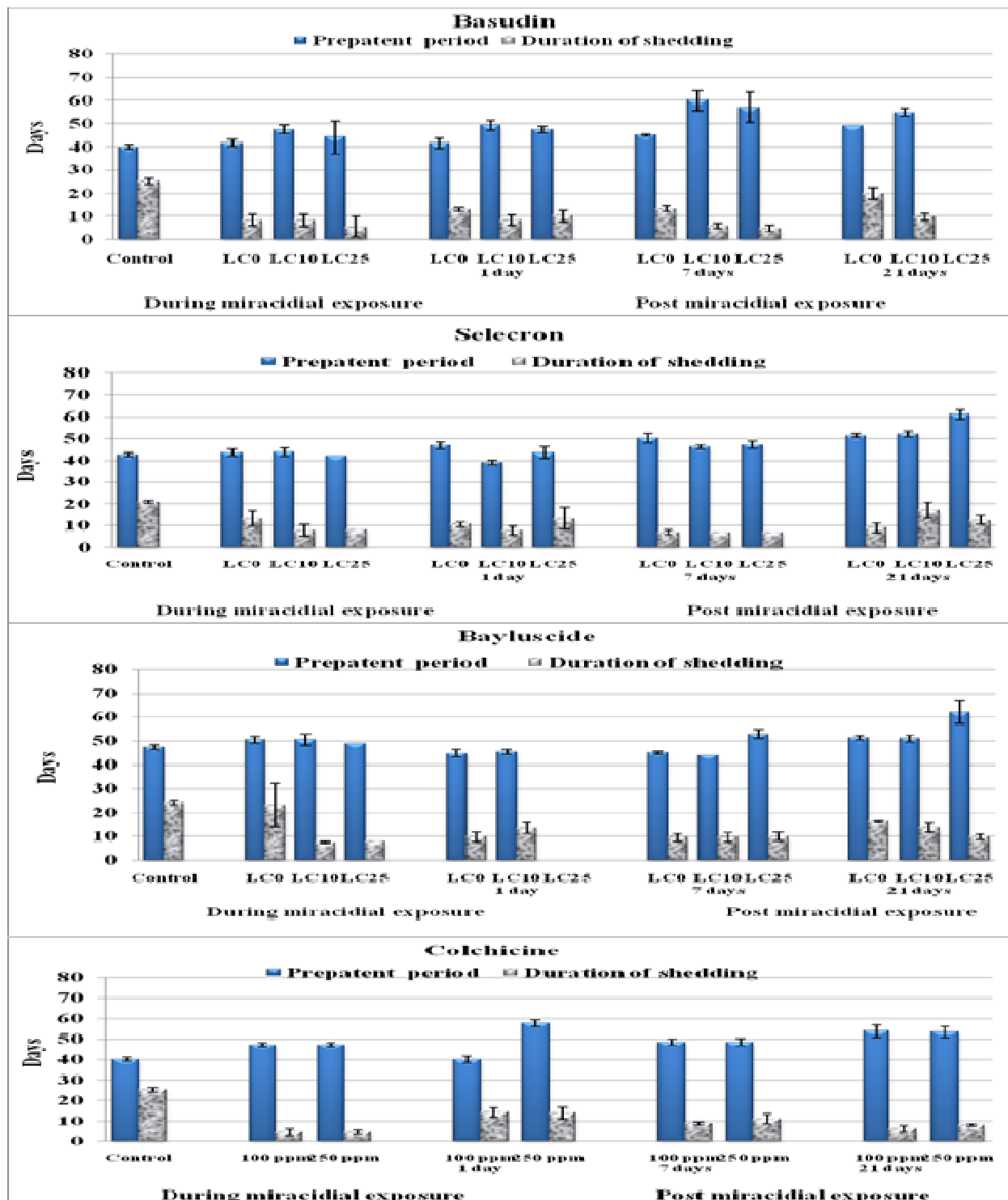


Figure 1. Effect of the compounds Basudin, Selecron, Bayluscide and Colchicine on prepatent period and duration of shedding of *Biomphalaria alexandrina* exposed to *Schistosoma mansoni*.

This study revealed a marked reduction of infection rates and cercarial production of snails treated with the tested

compounds in comparison with control groups. This was supported by the present records on disturbances in the

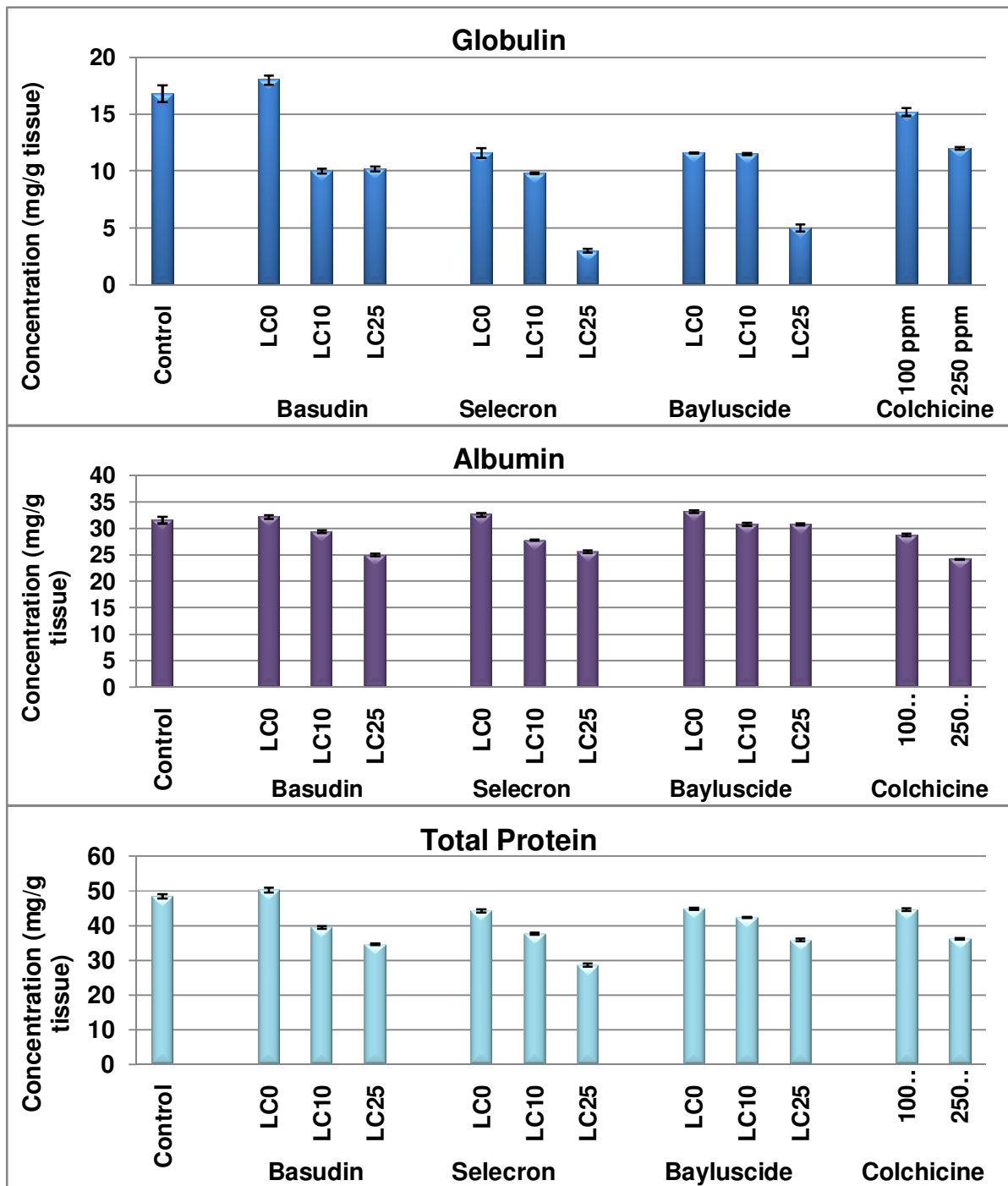


Figure 2. Effect of the compounds Basudin, Selecron, Bayluscide and Colchicine on globulin, albumin and total protein levels in tissue homogenate of *Biomphalaria alexandrina* snails.

activities of the enzymes AsT, AIT, AcP and AkP, also in the levels of total protein in tissues of *S. mansoni* infected snails treated with these agents, that may render their physiological processes unsuitable for the parasite development. In addition, the decrement of infection rates

of snails treated with the tested compounds during miracidial exposure may be due to the suppression of such agents on the infectivity and vitality of miracidia to penetrate the snails' skin and on their subsequent development within snails' tissues. Moreover, miracidial

Table 4. Effect of the compounds Basudin, Selecron, Bayluscide and Colchicine on biochemical parameters in tissue homogenate of *Biomphalaria alexandrina* snails (Mean±S.E).

Tested compound	Conc. ppm	AsT	AIT	AcP	AkP
		U/g protein	U/g protein	U/g protein	U/g protein
Control	0	2.8±0.16	4.0±0.19	28.4±0.16	3.7±0.22
	LC ₀	3.2±0.29	4.9±0.05	29±0.04	3.6±0.07
	LC ₁₀	4.1±0.13	6.3±0.22	28.6±0.05	5.9±0.18
Basudin	LC ₂₅	5.1±0.24	7.4±0.15*	17.7±0.16***	7.2±0.15*
	LC ₀	4.0±0.19	5.6±0.15	29.2±0.09	3.8±0.18
	LC ₁₀	4.4±0.16	6.5±0.20*	25.7±0.11*	3.9±0.05
Selecron	LC ₂₅	6.4±0.24*	9.3±0.11**	24.2±0.24*	5.6±0.05
	LC ₀	2.7±1.8	4.7±0.11	29.1±0.07	3.7±0.02
	LC ₁₀	4.9±1	7.2±0.16*	27.4±0.04	4.5±0.07
Bayluscide	LC ₂₅	5.3±0.24	7.9±0.20*	22.6±0.07***	5.3±0.04
	100	4.6±0.27	6.3±0.29	28.1±0.11	2.7±0.15
	250	5.2±0.33	8.2±0.35*	14.5±0.07***	2.1±0.04

Basudin: LC₀ (1.41 ppm), LC₁₀ (10.53) and LC₂₅ (12.25) - Selecron: LC₀ (0.402), LC₁₀(2.44) and LC₂₅ (3.11) – Bayluscide: LC₀ (0.02), LC₁₀ (0.07) and LC₂₅ (0.16). *, ** and *** = Significantly different from control at p< 0.05, p<0.01 and p<0.001, respectively.

penetration of snails' skin, in the presence of the tested compounds, damaged and ruptured snails' cells (Malek, 1980) causing more harmful stress and injury raising the snails death rates during the prepatent period, then reduced their survival and infection rates, shortened duration of cercarial shedding and cercarial production from infected snails under these stressors.

The present suppressive effect on cercarial production from infected treated snails could be due to the accumulation of the tested compounds in the snails' head-foot region specially for those treated during miracidial exposure and at early prepatent period, where mother sporocysts are still present (Malek, 1980) hence their subsequent developmental stages could be deteriorated leading to decrement in cercarial output and shortening the duration of cercarial shedding from these snails.

This was recorded by Mahmoud (2006a) on the insecticides Regent and Mimic, Esmail (2009) on the fungicide Topas and Youssef (2010) on the pesticides Match and Vertimec against infection of *B. alexandrina* snails with *S. mansoni*. Similar observations on infection of *B. truncatus* with *S. haematobium* post their exposure to the pesticides Chlorpyrifos and profenofos were recorded (Hasheesh and Mohamed, 2011). In addition, reduction in schistosomes cercarial output from infected-treated snails may be resulted from their active defense system. As, it was stated by Ataev and Coustau (1999) and Barbosa et al. (2006 a,b) that the success or failure of *B. glabrata* infection with trematodes depends on snails' humoral factors, mainly circulating hemocytes and plasma factors.

The present reduction of total protein and albumin concentrations in tissues of snails treated with the tested compounds could be a consequence of structural damages of their internal organs (Tolba et al., 1997). This was noticed by Esmail (2009) who attributed this reduction in *B. alexandrina* snails treated with the fungicide Topas to degeneration of the snail's ovotestis and digestive glands cellular structure. Meanwhile, the present study revealed an elevation in the activities of AsT, AIT and AkP enzymes in snails' tissues post treatment with LC₁₀ and LC₂₅ of Basudin, Selecron and Bayluscide in comparison with control groups. These observations are in agreement with those on *B. alexandrina* snails exposed to the pesticides Match and Vertimec (Youssef, 2010). However, Mahmoud and El-Sayed (2004) observed a decrease in the activity of AkP in hemolymph and tissues of *B. alexandrina* snails treated with LC₁₀ and LC₂₅ Niclosamide and attributed this to the acidity of snails' hemolymph which led to the rupture of their cells and severely damaged their organelles.

So, the present disturbances in the activities of the enzymes AsT, AIT, AcP and AkP in tissues of snails treated with the tested agents could be explained on the hypothesis of cells rupturing and injuries of different snails' organs, as well their suppressive effect on the nervous system of these snails. As, it was stated that the organophosphorus pesticides negatively affect the neurotransmitters in the nervous system of treated organisms (Van Cong et al., 2009).

From the foregoing data, it is concluded that low

concentrations of the pesticides Basudin and Selecron suppressed the vitality of *S. mansoni* larval stages (miracidia and cercariae) within a short exposure period. As well, they disturb the compatibility of *B. alexandrina* snails to *S. mansoni* through alterations of snails' metabolic processes. Therefore, accidental introduction of such pesticides to snails' habitats through plant pests control could negatively interfere with and/or prevent schistosomiasis transmission.

REFERENCES

- Ataev GL, Coustau C (1999). Cellular response to *Echinostoma caproni* infection in *Biomphalaria glabrata* strains selected for susceptibility/resistance. *Dev. Compar. Immunol.* 23(3):187-198.
- Babson LA (1965). Phenolphalein monophosphate methods for the determination of alkaline phosphatase. *Clin. Chem.* 11:789-796.
- Barbosa L, Caldeira RL, Carvalho OS, Vdighal TH, Jannotti-Passos LK, Coelho PM (2006a). Resistance of *S. mansoni* by transplantation of APO *Biomphalaria tenagophila*. *Parasitol. Immunol.* 28(5):209-212.
- Barbosa L, Shva LM, Coelho PM, Santos SR, Fortes-Dias CL (2006b). Primary culture of the region of amebocytes-producing organ of the snail *B. glabrata*, the intermediate host of *S. mansoni*. *Mem. Inst. Oswaldo. Cruz.* 101(6):639-643.
- Cabaj M, Toman T, Adamkovičová M, Massányi P, Šiška B, Lukáč N, Golian J (2010). Structural changes in the rat testis caused by diazinon and selenium. *Potravinárstvo* 4(2):8-16.
- Domas BT (1975). Standards for total serum protein assays. A collaborative study. *Clin. Chemist.* 21:1159-1166.
- El-Ansary A, Sammour EM, Soliman MS, Gawish FA (2001). In vivo attenuation of schistosome cercarial development and disturbance of egg-laying capacity in *Biomphalaria alexandrina* using sublethal concentrations of plant molluscicides. *J. Egypt Soc. Parasitol.* 31(3):657-669.
- El-khayat HMM, Abu-Zikri M (2004). Biochemical situation in *Biomphalaria alexandrina* infected with *Schistosoma mansoni* during twelve weeks post infection. *J. Egypt Ger. Soc. Zool.* 43(A):57-75.
- Esmail EA (2009). Biological and immunological studies on *Biomphalaria alexandrina* snails, the intermediate host of *Schistosoma mansoni* in Egypt. Ph.D. Thesis, Fac. Sci., Menoufia Univ. Egypt.
- Goldstein A (1964). *Biostatistics: An introductory text.* Macmillan, New York, p. 51.
- Gomes J, Dawodu AH, Lloyd O, Revitt DM, Anilal SV (1999). Hepatic injury and disturbed amino acid metabolism in mice following prolonged exposure to organophosphorus insecticides. *Hum. Exp. Toxicol.* 18(1):33-37.
- Gustafsson JEC (1976). Improved specificity of serum albumin determination and estimation of acute phase reactants by use of the bromocresol green reaction. *Clin. Chem.* 22:616-622.
- Hasheesh WS, Mohamed RT (2011). Bioassay of two pesticides on *Bulinus truncatus* snails with emphasis on some biological and histological parameters. *Pestic. Biochem. Physiol.* 100(1):1-6.
- Hira DR, Webbe G (1972). The effect of sublethal concentration of the molluscicide Triphenyl lead acetate on *Biomphalaria glabrata* (Say) and on the development of *Schistosoma mansoni* in the snail. *J. Helminthol.* 46:11-26.
- Kibiki GS, Drenth JP, Nagengast FM (2004). Hepatosplenic schistosomiasis: A Review. *East Afr. Med. J.* 81(9):480-485.
- Kreuger, J., 1999. Pesticides in the environment-Atmospheric deposition and transport to surface waters. Ph.D. Thesis, Swedish University of Agricultural Science, Uppsala, Sweden.
- Liang YS, Bruce JI, Body DA (1987). Laboratory cultivation of schistosome vector snails and maintenance of schistosome life cycle. *Proc. 1st Sino-Amer. Symp.* 1:34-48.
- Litchfield JT, Wilcoxon F (1949). A simplified method of evaluating dose-effect experiments. *J. Pharmacol. Exp. Therap.* 96:99-113.
- Mahmoud MB (2006a). Biological and histological impacts of the insecticides Regent and Mimic on *Biomphalaria alexandrina* snails. *Egypt. J. Zool.* 46:11-21.
- Mahmoud MB, El-Sayed KA (2004). Impact of Niclosamide on certain enzymes of *B. alexandrina* and some naturally associated snails. *Egypt. J. Zool.* 43:291-307.
- Malek EA (1980). Schistosomiasis. In: *Snail transmitted parasitic diseases.* Ed. CRC Press, Inc., Boca Raton, Florida, USA, pp. 179-307.
- Massoud J, Arafaa F, Chu KY (1973). Effect of Bayluscide (Bayer73) on the development of *Schistosoma haematobium* in *Bulinus truncatus*. *Bull. Soc. Pathol. Exot.* 66(4):544-547.
- Molad Y (2002). Update on colchicine and its mechanism of action. *Curr. Rheumatol. Rep.* 4:252-256.
- Moss DW (1984). *Methods of enzymatic analysis,* Bergmeyer, H.U. (ed.), Verlag - Chemi. Weinheim 4:92-106.
- Reitman S, Frankel S (1957). A colorimetric method for determination of serum glutamic oxaloacetic and glutamic pyruvic transaminase. *Am. J. Clin. Pathol.* 28:56-61.
- Salih EMA (2010). Toxic Effect of Dimethoate and Diazinon on the Biochemical and Hematological Parameters in Male Rabbits. *J. Biol. Sci.* 3(2):77-82.
- Southwood TRE (1978). *Ecological methods.* Halsted press, Chapman and Hall, London, p. 524.
- Tchoun-Wou PB, Englande JA, Malek EA (1991). Toxicity evaluation of Bayluscide and malathion to three developmental stages of freshwater snails. *Arch. Environ. Contam. Toxicol.* 21(3):351-358.
- Tolba MR, Mohamed B, Mohamed M (1997). Effect of some heavy metals on respiration, mean enzyme activity and total protein of the pulmonate snails *B. alexandrina* and *B. truncatus*. *J. Egypt. Ger. Soc. Zool.* 24(D):17-35.
- Tomlin CDS (1997). *The pesticide manual,* 11th Ed (ISBN 1 901396 11 8). British Crop Protection Council, UK.
- Van Cong N, Phuong NT, Bayley M (2006). Sensitivity of brain cholinesterase activity to Diazinon (Basudin 50EC) and Fenobucarb (Bassa 50EC) insecticides in the air-breathing fish *Channa striata* (Bolch, 1793). *J. Environ. Toxicol. Chem.* 25(5):1418-1425.
- Van Cong N, Phuong NT, Bayley M (2009). Effects of repeated exposure of diazinon on cholinesterase activity and growth in snakehead fish (*Channa striata*). *Ecotoxicol. Environ. Safety* 72(3):699-703.
- White BA, Erickson MM, Stevens SC (1970). Determination of sGoT and sGPT enzymes; in *Chemistry for Medical Technologists* pp. 293-296.
- World Health Organization (1965). *Molluscicide screening and evaluation.* Bull. WHO 33:567-581.
- Yousef MI, Awad TI, Mohamed EH (2006). Deltamethrin-induced oxidative damage and biochemical alterations in rat and its attenuation by Vitamin E. *Toxicology* 29(3):240-247.
- Youssef AA (2010). Studies on the impact of some pesticides and Egyptian plants on some biological and physiological parameters of *Biomphalaria alexandrina* snails and their susceptibility to infection with *Schistosoma mansoni* miracidia. M.Sc. Thesis, Faculty of science, Al-Azhar University, Egypt.