Egyptian Journal of Aquatic Biology & Fisheries
Zoology Department, Faculty of Science,
Ain Shams University, Cairo, Egypt.
ISSN 1110 - 6131 Vol. 22(3): 71- 87 (2018)
ejabf.journals.ekb.eg

Potential role of rice cultivation and fish aquaculture in the transmission of schistosomiasis and the effect of pesticides and fertilizers used in rice cultivation on the snail *Biomphalaria alexandrina* and schistosome larval stages

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ARTICLE INFO

Article History:
Received: May12, 2018
Accepted:June 30, 2018
Available online:J uly 2018

Keywords:
Schistosomiasis
B. alexandrina
B. truncatus
Rice cultivation
Aquaculture
Pesticides
fertilizers

ABSTRACT

The present investigation aimed to study the potential role of rice cultivation and fish aquaculture in the transmission of schistosomiasis in Kafr El-Sheikh governorate. This study accomplished in four non-governmental rice fields; three non- governmental fish aquaculture and one governmental of each about one year. It was found that *B. alexandrina* established in all watercourses next to nongovernmental farms in the whole study, while *B. truncatus* was observed in certain watercourses next to non-governmental farms. The toxicity of six pesticides (lampada, sylian, saturn, karasay, mancozeb and machete and two fertilizers urea and mixed nitrate) which used in rice cultivation against *B. alexandrina* and their egg masses and schistosome miracidia and cercariae showed that all pesticides had strong molluscicidal properties and are far toxic than fertilizers. Lampada and karasay were the strongest pesticides against snails where their LC50 & LC_{90} were 0.8 & 1.0 and 0.7 & 1.1 ppm, respectively. Also, fertilizers LC_{50} & LC_{90} were 2754 & 4092 ppm for mixed nitrate and 9636 & 15937 ppm for urea. Effect of LC_{25} and LC_{50} of pesticides and fertilizers on mortality of *B. alexandrina* eggs explored that fertilizer mixed nitrate more potent effect on the eggs that killed 97.6% and 98.2%, respectively. Followed by Pesticides mancozeb and machete witch caused 94.7 & 95.9% and 94.2 and 97.9% mortality, respectively. The current data indicated that death rates of *S.mansoni* miracidia and cercariae were started after minutes of exposure to LC_5 of both pesticides and fertilizers. It is concluded that accidental introduction of such pesticides to snails' habitats through plant pests control, low concentrations of the pesticide mancozeb and mixed nitrate inhibited the hatchability of *B. alexandrina* eggs, also suppressed vitality of *S. mansoni* larval stages within a short exposure period. As well; they disturb the compatibility of *B. alexandrina* snails to *S. mansoni* miracidia.

INTRODUCTION

In Egypt, schistosomiasis is a major public health problem due to its high prevalence and morbidity among the Egyptians, especially among the rural populations.

People are infected by contact with water used in normal daily activities such as personal or domestic hygiene and swimming, or by professional activities such as fishing, rice cultivation and irrigation (Abdel-Wahab *et al*., 2000; Gabr *et al*., 2000 and El-Baz *et al*., 2003). Kafr El-Sheikh governorate is one of the highest rice-producer among Egyptian governorates. During rice cultivation, due to the farmers' direct contact with the irrigation water, they infected with some water related diseases, the most important of which is schistosomiasis (Joshi and Phil 2002). Pesticides are applied widely to protect plants from diseases, weeds, fungal and insect damage. Although the use of pesticides has resulted in increased crop production and other benefits, it has raised concerns about potential adverse effects on the environment and aquatic life (Aktar *et al*., 2009 and Delfina *et al.*, 2016). In addition to fertilizers used for improving, increasing growth and yield of crops may reach snails in canals via drainage system (Ragab and Shokry, 2006). These Fertilizers have been repeatedly to be effective in the presence of schistosome snails in particular medium (Tantawy, 2006). The toxic effect of pesticides and fertilizers on the survival of schistosome miracidia and cercariae are proven in several studies (Mostafa, 2004 & 2006; El-Emam *et al.*, 2008 and Ismail, 2009) concluding that cercariae is more sensitive than miracidia.

The main objective of the present study was to assess schistosomiasis transmission next to rice farms and fish aquacultures, also, to compare between transmission in governmental and non-governmental farms. In addition, to investigate the toxicity of six pesticides (lampada, sylian, saturn, karasay, mancozeb and machete) and two fertilizers (urea and mixed nitrate) which applied in the cultivation of rice in Egypt against *Biomphalaria alexandrina* snails and the potency of these compounds on S. mansoni miracidia and cercariae .

MATERIALS AND METHODS

The study area and Filed study:

The investigated farms were chosen in Kafr El-Sheikh governorate to be irrigated by the most active schistosome transmitting watercourses as five rice farms were irrigated with the following water courses, El-Alawy canal in Fuwwah center, Om Youssef drain in Desouk center, Menyet Al-Morshed canal in Mottobis center, Rewenah canal in Kafr ElSheikh center (the Governmental) and Kirodrain drain in Biyala center. The selected fish aquacultures were four, two of them are in Baltim center; the Borollos and El-Maadia Gebli aquacultures which are located in El-Borollos lake and Kotshenar drain, respectively. The other two aquacultures are in El-Hamoul center; El-Tharwa and El-Hamoul aquacultures which are supplied with the water courses, agricultural drain no.6 and El-Tashween drain (Fig.1). The Study was conducted in the period between May-September, 2011 in rice farms and December, 2010-December, 2011 in fish aquacultures. The field survey has been done by the collection of *B. alexandrina* and *Bulinus truncatus* snails from the watercourses under study using a standard dip net (Yousif et al., 1992). Dissolved oxygen was measured by dissolved oxygen meter electrode (HANNA HI 9146). Water temperatures and conductivity were recorded using temperature conductivity meter (HANNA instrument, HI 9635). Also, pH was measured by pH meter electrode (HI 9124 and HI 9125). Water samples were taken from each study site.

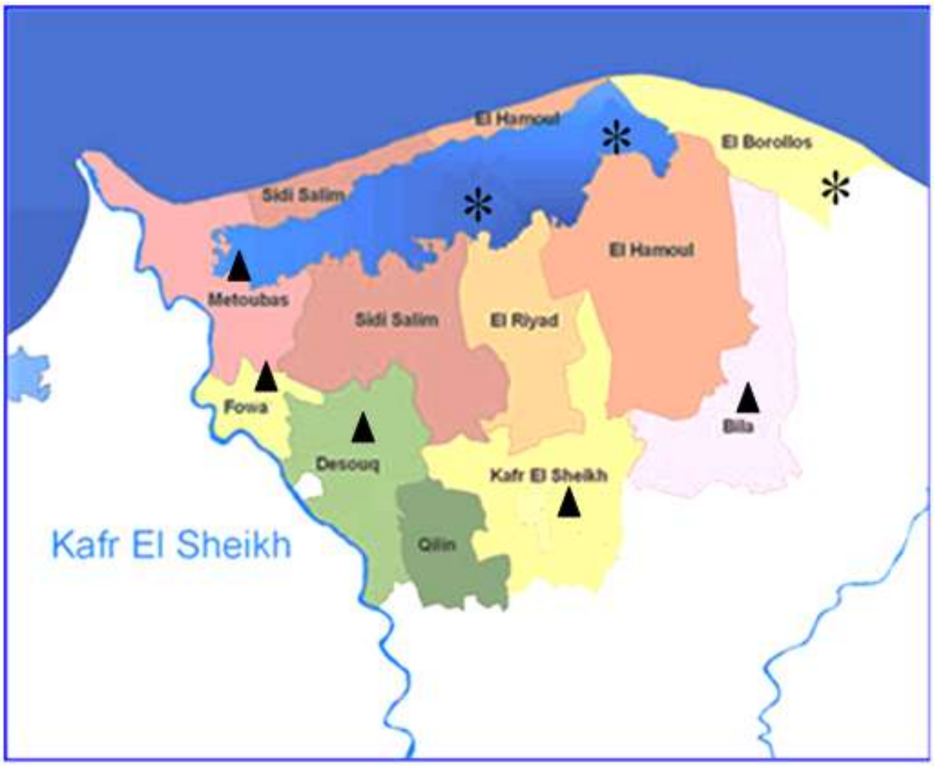

Fig. 1: study area in Kafr El Sheikh governorate (▲ rice farming and* fish aquacultures).

Laboratory study:

At the laboratory, *B. alexandrina* and *B. truncatus* were counted and examined for their natural infection individually at weak intervals for one month according to Favre *et al.* (1995). Water samples were filtrated and prepared for measuring the concentration of Cd, Pb, Cu, Fe, Ni, Na, K and Ca by the atomic absorption spectrophotometer (AVANTA).

Snails:

B. alexandrina were collected from the irrigation schemes in Kafr El-Sheikh governorate and transferred to the laboratory where they kept in plastic aquaria (35×25×10 cm) containing dechlorinated tap water (10 snails/aquarium) and fed by fresh leaves of lettuce. Snails maintained as such for about 1 month before being used in toxicity experiments.

Toxicity test:

Pesticides (lampada, sylian, sturn, karasay, mancozeb and machete) and fertilizers (urea and mixed nitrate) were obtained from Ministry of Agriculture and Lands Reclamation, Rice Research and Training Center, Sakha, Kafr El-Sheikh (Field Crop Research Institute, Agriculture Research Center). To evaluate the efficacy of these tested chemicals against adult snails a series of concentrations were prepared from stock solution of 1000 ppm for each of the examined pesticides and fertilizers. Three replicates were used, ten snails (6-8 mm)/L, for each concentration. Exposure and recovery periods were 24 h each; at 25±1°C. For each test, 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). Groups of *B. alexandarina*, each of 15 snails were maintained in plastic aquaria containing dechlorinated tap water. Thin plastic sheets for egg deposition were provided in the aquaria and boiled lettuce leaves and fish food were added twice a week. The eggs were collected daily and maintained in separate aquaria (ElKhayat, 2008).

Quantification of eggs and egg-masses:

The determination of ovicidal activity of pesticides and fertilizers against *B. alexandrina* eggs was made according to El-Khayat *et al.*, (2003a). Three concentrations (LC_{25}, LC_{50} and LC_{90}) as assessed against snails in three replicates from each tested material were prepared in 100 ml of dechlorinated tap water. So, another set with dechlorinated tap water as a control. The egg clutches of the same age (3day) were collected and examined under a binocular microscope to confirm viability according to Abdel-Kader *et al.* (2005). Five egg clutches were added to each concentration and control after their embryos number recording. After 24h of exposure, the egg clutches were washed thoroughly in dechlorinated water and kept for another 24h as a recovery period. Surviving and dead embryos in the exposed egg clutches were counted then the mortality percentages were registered and corrected according to Abbott's formula (1925). In addition, the egg masses treated by previous concentrations were collected and maintained for hatchability according to Tantawy *et al.* (2008).

Cercariael and miracidial activities:

S. mansoni cercariae and ova were obtained from the Schistosome Biological Supply Center (SBSC), Theodor Bilharz Research Institute (TBRI). The freshly hatched miracidia and cercariae were used to examine the miracidial and cercariael activity against the tested pesticides and fertilizers. Three replicates were used for each concentration and control groups. Each replicate of 100 cercariae or miracidia were exposed to 10 ml of LC_5, LC_{10} and LC_{25} as assessed against snails. Ten ml of declorinated water containing 100 cercariae or miracidia were used as control (Hasheesh and Mohamed, 2011). The mortality was determined at different time intervals; 3/4, 1, 2, 3 & 4 h, then a percentage of cumulative mortality of miracidia and cercariae were determined.

Statistical analysis:

All analyses were performed using SPSS version 19 (SPSS, Inc., Chicago, IL) the analysis included the calculation of the mean value, standard deviation and test for difference between proportion (2 groups) to examine correlation between the infestation percentage of each of *B. alexandrina* & *B. truncatus* in different centers. P values <0.05 were considered statistically significant. Also, one-way analysis of variance (ANOVA) was used to compare physicochemical parameters between snails’ habitats in governmental and non-governmental farms.

RESULTS

Distribution and density of *B. alexandrina* and *B. truncatus* in five water courses next to rice farms are summarized in Table (1). *B. alexandrina* snails were established in the four water courses next to non-governmental farms. The highest infestation percentage was in Fuwwah and Desouk centers (100%), followed by Mottobis (80%). In spite of Fuwwah and Desouk centers were characterized by the highest snail density (34.4 and 21.6 snails/site, respectively) they showed the lower density of naturally infected snails (0.6, 0.4 and 4.0 snails/site, respectively). Although Biyala center showed lower snail density (3±1.871 snails/site), a high percentage of infection (13.3%) was recorded. On the other hand, Mottobis and Kafr El-Sheikh centers were free from naturally infected snails. In the case of *B. truncatus*, snails were found in Mottobis center only, with infestation percentage 40% (the snail density was 1 snail/ site). Naturally infected *B. truncatus* snails were observed in Mottobis center (20%) with adensity mean of 0.2 snail/ site. In contrast, Kafr El – sheikh center (governmental farm) was free from *B. alexandrina* and *B. truncatus* snails. Comparing both snail species in Kafr El–Sheikh governorate revealed that the density of *Biomphalaria* was about 80 times than *Bulinus*, while the percentage of naturally infected *B. truncatus* snails was higher than *B. alexandrina* snails (Table 1). Statistical analysis showed that the infestation pattern of *B. alexandrina* was highly significant in Fuwwah center (P≤ 0.005) and in El-Reyad (P≤ 0.01) but it was significant in Mottobis and Biyala (P≤0.05).

Table 1: The distribution and population density of *B. alexandrina* and *B. truncatus* in the selected water courses in rice farms

Centers	**Fuwwah**		**Desouk**		**Mottobis**		**Kafr El-Sheikh**		**Biyala**	
Watercourses	**El-Alawy canal**		**Om Youssef drain**		**Menyet Al-Morshed canal**		**Rewenah canal**		**Kirodrain drain**	
Snails Items	***B.a***	***B.t***	***B.a.***	***B.t***	***B.a.***	***B.t***	***B.a.***	***B.t***	***B.a.***	***B.t***
No. of infested sites	5	0	5	0	1	2	0	0	4	0
% of Infestation	100%	0%	100%	0	20%	40%	0	0	80%	0
No.of transmission sites	3	0	2	0	0	1	0	0	2	0
% of infestation of transmission sites	60%	0%	40%	0	0	20%	0	0	40%	0
Total no. of Snails	172	0	108	0	2	5	0	0	15	0
Snail density (snail/site ±SD)	34.4 ±8.905	0	21.6 ±7.266	0	0.4 ±0.894	1 ±1.414	0	0	3 ±1.871	0
% of natural infection	1.7%	0	1.9%	0	0	20%	0	0	13.3%	0

B.a: Biomphalaria alexandrina, B.t: Bulinus truncatus

B. alexandrina and *B. truncatus* density of distribution in four water courses next to fish aquacultures was represented in Table (2). *B. alexandrina* snails were found established in the three watercourses El-Borolos Lake, Kotshenar drain, El-Tashween canal. The highest percentage of infestation was 100% in El-Borolos Lake and Kotshenar drain, while the lowest was in El-Tashween canal being 30.8%. Kotshenar drain and El-Borolos Lake were characterized by the highest snail density (36.385 and 20.846 snails/site, respectively). Kotshenar drain was the mostly infested with naturally infected *B. alexandrina* (38.5%). Although El-Tashween canal showed the lowest value of snail infestation it had the highest percentage of natural infection (25%). El-Borolos Lake recorded the lower percentage of natural infection (1.8%) in spite of characterizing by the highest snail density. In case of *B. truncatus* snails, the high infestation percentage was in Kotshenar drain and Agriculture drain no.6 (30.8%) and snail density was 0.615 and 0.923 snail/site, respectively. Naturally infected *B. truncatus* snails in Kotshenar drain were 12.5%. In contrast, other watercourses were free from naturally infected snails. Comparing both snail species in watercourses next to fish aquaculture revealed that the density of *Biomphalaria* was approximately 40 times that of *Bulinus*, but the percentage of naturally infected snails was higher among *B. alexandrina* snails (25%) than *B. truncatus* snails (12.5%).

Table 2: The distribution and population density of *B. alexandrina* and *B. truncatus* in selected fish aquacultures under investigation.

Centers	Baltim				El-Hamoul			
Aquacultures	El-Borollos		El-Maadia Gebli		El-Tharwa		El-Hamoul	
Watercourses	El-Borollos lake		Kotshenar drain		Agricultural drain no. 6		El-Tashween	
Snails	*B.a*	*B.t*	*B.a*	*B.t*	*B.a*	*B.t*	*B.a*	*B.t*
No. of infested sites	13	1	13	4	0	4	4	0
% of Infestation	100%f	7.7%	100%	30.8%	0	30.8%	30.8%	0
No. of transmission sites	4	0	5	1	0	0	1	0
% of infestation of transmission sites	30.8%	0%	38.5%	7.7%	0	0	7.7%	0
Total no. of Snails	271	2	473	8	0	12	4	0
Snail density (snail/site ±SD)	20.846 ±13.297	0.154 ±0.555	36.385 ±15.446	0.615 ±1.121	0	0.923 ±1.605	0.308 ±0.630	0
% of naturally infected snails	1.8%	0	8.0%	12.5%	0	0	25%	0

B.a: Biomphalaria alexandrina, B.t: Bulinus truncatus

Physico-chemical parameters in five water courses next to rice farms are presented in Table (3). Results showed that temperature, PH, Conductivity and D.O in all different examined watercourses were approximately the same ranges.

Table 3: Physiochemical parameters expressed by mean and SD in rice farms at Kafr El-Sheikh governorate.

Centers	Fuwwah	Desouk	Mottobis	Kafr El-Sheik	Biyala
Watercourses Items	El-Alawy Canal	Om Youssef Drain	Menyet Al-Morshed Canal	Rewenah Canal	Kiro Drain
Temperature	27.0 ^{C} ±2.23	26.4 ^{C} ±3.36	29.2 ^{BC} ±3.70	29.2 ^{BC} ±4.32	28.4 ^{BC} ±2.70
Conductivity	1052.0 ^{ab} ±243.14	1216.0^{a} ±359.35	732.0 ^{c} ±129.35	907.6 ^{bc} ±86.13	1018.2 ^{abc} ±292.24
pH	8.2 ^{ABC} ±0.42	7.8 ^{C} ±0.21	8.0 ^{BC} ±0.31	8.0 ^{BC} ±0.00	7.9 ^{BC} ±0.31
Dissolved oxygen	2.9 ^{b} ±1.68	3.2 ^{b} ±2.04	4.8 ^{ab} ±0.44	5.4 ^{ab} ±2.96	8.1 ^{a} ±4.90
Cd	0.48 ^{BC} ±0.38	0.66 ^{Bc} ±0.44	1.30 ^{A} ±0.75	0.15 ^{C} ±0.13	0.15 ^{C} ±0.11
Pb	17.45^{e} ±10.39	17.46^{e} ±12.37	8.24^{e} ±2.17	16.77^{e} ±15.83	14.42^{e} ±10.15
Fe	59.0^{e} ±2.01	70.84^{e} ±25.83	50.34^{e} ±35.97	41.89^{e} ±20.92	57.21^{e} ±32.51
Cu	55.16^{e} ±18.31	62.18^{e} ±22.17	48.51^{e} ±14.23	47.79^{e} ±27.64	49.76^{e} ±33.76
Na	119.47 ^{B} ±99.16	97.07 ^{B} ±5.27	76.85 ^{B} ±22.35	152.83 ^{B} ±105.57	482.59^{A} ±363.55
K	15.70^{e} ±2.23	20.42^{e} ±10.23	16.30^{e} ±4.81	21.59^{e} ±14.72	25.80^{e} ±11.06
Ca	330.59^{e} ±61.37	321.42^{e} ±81.63	253.73^{e} ±41.26	390.89^{e} ±127.78	294.14^{e} ±77.10

a, b Means having different letters exponents among rows are significantly different($P \leq 0.05$).A, B Means having different letters exponents among rows are significantly different ($P \leq 0.01$). (According to Duncan's multiple range- test. e Means having letter exponents among rows are non- significant.

Also, the concentrations of Cd, Pb, Fe, Cu, K and Na were approximately the same or fluctuated in narrow ranges. In contrast, Ca concentration increased in all watercourses examined in comparison to other metals.

Table (4) cleared that temperature, pH, conductivity and D.O in all different examined watercourses recorded approximately the same ranges, while D.O concentration in El-Hamoul was higher (7.2ppm). The concentration of Pb, Fe, Cu, K and Ca in different examined watercourses were approximately the same or fluctuated in narrow ranges, while K and Pb recorded high concentration (71.085 and 49.619 ppm, respectively) in agricultural drain no. 6. Cd concentration in El-Borollos lake was high (34.18 ppb ± 24.77) while it waslow in El-Hamoul. The Na concentration recorded three levels, high concentration in El-Borollos lake (100.89ppm ± 52.33) and low in Agricultural drain no.6 (15.25 ppm ± 8.54).

Table 4: The Physico-chemical characteristics of the watercourses next to fish aquacultures at Kafr El-Sheikh governorate.

Centers	**Baltim**		**El-Hamoul**	
	El-Borollos lake	**Kotshenar drain**	**El-Tharwa Agriculture. N.6**	**El-Hamoul El-Tashween**
Temperature	23.5^{e} ±5.060	24.5^{e} ±5.6	24.6^{e} ±8.6	26.3^{e} ±5.2
Conductivity	1787.1^{ab} ±1717.0	2215.8^{a} ±1577.6	1100.7^{b} ±435.2	1458.1^{ab} ±718.8
pH	7.9^{b} ±0.234	8.1^{b} ±.4	7.9^{b} ±2.4	8.4^{ab} ±0.6
D.O	4.8^{ab} ±3.5	5.1^{ab} ±3.2	4.4^{ab} ±3.8	7.2^{a} ±4.9
Cd (ppb)	34.18^{A} ±24.981	8.11^{B} ±6.84	4.28^{B} ±4.07	5.52^{B} ±3.497
Pb (ppb)	9.80^{B} ±7.48	1.24^{C} ±1.01	0.37^{C} ±.24	18.81^{A} ±12.087
Fe (ppb)	38.49^{e} ±24.98	33.460^{e} ±28.44	25.12^{e} ±22.81	18.73^{e} ±6.97
Cu (ppb)	46.060^{ab} ±20.806	41.26^{b} ±21.77	37.70^{b} ±11.43	60.22^{a} ±17.81
Na (ppm)	100.98^{A} ±52.333	42.67^{B} ±26.570	15.25^{C} ±8.45	45.25^{B} ±18.982
K (ppm)	25.09^{A} ±9.64	15.16^{BC} ±8.18	15.25^{C} ±8.45	17.28^{B} ±6.97
Ca (ppm)	86.37^{e} ±51.11	109.35^{a} ±19.679	86.15^{e} ±17.04	111.64^{a} ±16.62

a, b Means having different letters exponents among rows are significantly different ($P \leq 0.05$).A, B Means having different letters exponents among rows are significantly different ($P \leq 0.01$). (According to Duncan's multiple range- test. e Means having letter exponents among rows are non- significant.

Comparison between Governmental and non-Governmental farms:-

Table (5) explained that *B. alexandrina* snails were established in rice farms and fish aquacultures (Non-governmental) with infestation percentage of 60% and 76.9%, respectively. Non-governmental sites were characterized by the highest snail density (19.077 snails/ site in fish aquacultures and 14.85 snails/ site in Rice farms). Rice farms (non-governmental) were the mostly infested with *B.alexandrina* (46.7%) while governmental rice farms and fish aquacultures were free from *B. alexandrina*. Non-governmental fish aquacultures and rice farms showed natural infection of 5.9% and 2.4% respectively). The infestation percentages of *B. truncatus* snails found in non-governmental rice farms and fish aquacultures were 10% and 2.6%, respectively.

Naturally infected *B. truncatus* snails were observed in non-governmental rice farms and fish aquacultures were 20% and 10%, respectively. Comparing both snail species in non-governmental rice farms and fish aquacultures revealed that the density of *Biomphalaria* was higher in fish aquacultures than that in rice farms.In rice farms (non-govenmental the percentage of naturally infected snails was higher among *B. truncatus* snails (20%) than *B. alexandrina* snails (2.4%).

Table 5: The distribution and population density of *B. alexandrina* and *B. truncatus* in governmental and non-governmental rice farms and fish aquacultures.

Farms	Rice farm				Fish Aquaculture			
	governmental		non-governmental		governmental		non-governmental	
Snails Items	***B.a***	***B.t***	***B.a***	***B.t***	***B.a***	***B.t***	***B.a***	***B.t***
No. of Infested sites	0	0	15	2	0	0	30	1
% of Infestation	0	0	60%	10%	0	0	76.9%	2.6%
Total no. of Snails	0	0	297	5	0	0	748	10
Snail density (snail/site)	0	0	14.85	0.25	0	0	19.077	0.26
No. of transmission sites	0	0	7	1	0	0	10	1
% of infestation of transmission sites	0	0	35%	5%	0	0	25.6%	2.7%
No. of Infected snails	0	0	7	1	0	0	44	1
Infected snail density (infected snails/site ±SD)	0	0	0.35	0.05	0	0	1.1	0.026
% of naturally infected snails	0	0	2.4%	20%	0	0	5.9%	10%

B.a: Biomphalaria alexandrina, B.t: Bulinus truncatus

Comparing between rice farms and fish aquaculture (governmental, non-governmental) in Table (6) indicated that all physical parameters recorded were approximately the same or fluctuated in narrow ranges, with the exception in fish aquacultures (non-governmental) for conductivity was1458.1±718.81 µmohm / Cm.

Table 6: Comparison of physicochemical parameters in governmental and non-governmental rice farms and fish aquacultures

Items (Mean ± SE)	Rice farm		Fish Aquaculture	
	governmental	**non-governmental**	**governmental**	**non-governmental**
Conductivity	907.6 ± 86.13^{b}	1004.6 ± 305.10^{b}	1192.4 ± 295.43^{ab}	1458.1 ± 718.81^{a}
pH	8.0 ± 0.00^{B}	8.0 ± 0.32^{B}	8.5 ± 0.38^{A}	8.4 ± 0.58^{A}
D.O.	5.4 ± 2.96^{e}	4.8 ± 3.30^{e}	4.8 ± 3.67^{e}	7.2 ± 4.89^{e}
Cd (ppb)	0.15 ± 0.12^{B}	0.7 ± 0.60^{B}	4.3 ± 4.07^{A}	5.3 ± 3.49^{A}
Pb (ppb)	16.8 ± 15.82^{A}	14.4 ± 9.42^{A}	0.4 ± 0.24^{B}	18.8 ± 12.08^{A}
Fe (ppb)	41.9 ± 20.92^{AB}	59.3 ± 25.74^{A}	25.1 ± 22.81^{BC}	18.7 ± 6.97^{C}
Cu (ppb)	47.8 ± 27.63^{ab}	53.9 ± 21.55^{ab}	37.7 ± 11.43^{b}	60.2 ± 17.81^{a}
K (ppm)	21.6 ± 14.72^{a}	19.6 ± 8.26^{a}	11.1 ± 3.56^{b}	17.3 ± 6.97^{ab}
Ca (ppm)	390.9 ± 127.77^{A}	299.9 ± 74.75^{B}	86.2 ± 17.04^{C}	111.6 ± 16.62^{C}

$^{a, b}$ Means having different letters exponents among rows are significantly different ($P \leq 0.05$).
$^{A, B}$ Means having different letters exponents among rows are significantly different ($P \leq 0.01$).

Also, the concentrations of Cd, Pb, Fe, Cu, K and Cd in rice farms and fish aquacultures were approximately fluctuated in narrow concentration. On the other hand, there was increasing in the concentration of Ca in rice farms (governmental and non-governmental) 390.9and 299.9,respectively. while in fish aquacultures 86.2 and 111.6 (governmental and non-governmental, respectively) . Statistical analysis of physicochemical parameters between governmental and non-governmental rice farms and fish aquacultures explains that, there was high significant difference between governmental and non-governmental for rice farms and fish aquacultures in PH, Cd, Pb, Fe, Ca (P<0.01), while there was significant in Conductivity, Cu & K (P<0.05).

The toxicity of the selected common agriculture six pesticides; lampada, sylian, saturn, karasay, mancozeb and machete and two fertilizers urea and mixed nitrate were assessed against adult *B. alexandrina* snails (Table7). LC_{90} values of the tested Pesticides and fertilizers were 1.0 ppm for lampada, 10 ppm for sylian, 8.5 ppm for saturn, 383.2 ppm for mancozeb, 33.2 ppm for machete, 1.1 ppm for karasay, 15937 ppm for urea and 4092 ppm for mixed nitrate, while it was 0.21 ppm for bayluscide which was used as a control.

Table 7: Influence of pesticides and fertilizers against adult *Biomphalaria alexandrina snails*

Pesticides / Fertilizer	LC_0	LC_5	LC_{10}	LC_{25}	LC_{50}	Upper limit	Lower limit	LC_{90}	Slope	X^2	
										observed	tabled
Lampada 5%	0.3	0.45	o.5	0.6	0.8	0.9	0.7	1.0	1.3	10.05	11.070
Sylian 72% EC	2.2	4.3	4.5	5.2	6.3	7.2	5.5	10	1.4	5.660	5.941
Saturn 50 % EC	2.7	4	4.4	5.0	6.0	6.7	5.4	8.5	1.3	6.720	9.488
Mancozeb 80% WP	21.0	56.0	70.0	105.0	191.0	247.0	147.0	383.2	1.9	8.640	9.488
Machete 60% EC	1.8	4.0	6.2	9.0	14.7	18.97	11.39	33.2	1.9	6.002	9.488
Karasay 3.5% EC	0.25	0.41	0.45	0.56	0.7	0.9	0.5	1.1	1.4	1.445	9.488
Urea	2800	4749	5734	7320	9636	11756	7898	15937	1.5	0.695	9.488
Mixed nitrate	1000	1537	1816	2050	2754	3255	2330	4092	1.3965	4.705	7.815
Bayluscide	0.1	0.13	0.14	0.15	0.17	0.2	0.16	0.21	1.2	1.760	7.815

The effect of LC_{25} and LC_{50} of pesticides and fertilizers against *B. alexandrina* eggs (Table 8) revealed that mixed nitrate had the more potent effect on the eggs which killed 97.6% and 98.2%, respectively, followed by mancozeb (94.7% & 95.9%, respectively) and machete (94.2% and 97.9%, respectively). Also, LC_{90} of mixed nitrate ; mancozeb and machete were caused 100% mortality of eggs.

Table 8: Effect of pesticides and fertilizers on mortality of B. alexandrina eggs

Mortality %	Dead	Examined	Concentrations	Pesticide /Fertilizer
9.5 %	4	42	LC_{25} (0.56 ppm)	**Karasay**
16 %	8	49	LC_{50} (0.74 ppm)	
50 %	14	28	LC_{90} (1.10 ppm)	
3.4 %	3	88	LC_{25} (5.20 ppm)	**Sylian**
19.4 %	14	72	LC_{50} (6.30 ppm)	
44 %	26	58	LC_{90} (8.50 ppm)	
94.2 %	65	69	LC_{25} (5.10 ppm)	**Machete**
97.9 %	47	48	LC_{50} (14.80 ppm)	
100 %	56	56	LC_{90} (33.20 ppm)	
11.3%	8	71	LC_{25} (4.70 ppm)	**Saturn**
16.1%	10	62	LC_{50} (6.00 ppm)	
94.7 %	54	57	LC_{90} (8.50 ppm)	
2.3 %	2	87	LC_{25} (0.60 ppm)	**Lampada**
5.6 %	4	71	LC_{50} (0.80 ppm)	
14.8 %	9	61	LC_{90} (1.00 ppm)	
94.7 %	36	38	LC_{25} (89.50 ppm)	**Mancozeb**
95.3 %	41	43	LC_{50} (190.80 ppm)	
100 %	37	37	LC_{90} (383.20 ppm)	
3.5 %	3	85	LC_{25} (6319.90 ppm)	**Urea**
5.3 %	3	56	LC_{50} (9635.90 ppm)	
25 %	9	36	LC_{90} (15973.00 ppm)	
97.6 %	82	84	LC_{25} (2049.70 ppm)	**Mixed nitrate**
98.2 %	60	61	LC_{50}(2753.80 ppm)	
100 %	**34**	**34**	**LC_{90} (4091.70 ppm)**	

In addition, the effect of pesticides and fertilizers on hatchability of *B. alexandrina* egg (Fig 2) showed that the hatchability rate was the highest when exposed to LC_{25} of karasay (81%), while it was reduced to 50% by exposure to LC_{90}. Meanwhile, hatchability rate was 67% and 22.4 % by exposure to LC25 and LC_{90} of Sylian and decreased to 1.4% by the exposure to LC_{25} of machete , whereas there was no hatchability recorded with the exposure to LC_{90}. In the meantime hatchability rate was 11.3% for LC_{25} of saturn, meanwhile, it was decreased to 1.8% by the exposure to LC_{90}. In contrast, hatchability rate was the highest being 95.4% by the exposure to LC_{25} lampada, while it was 75.4 % when exposed to LC_{90}. On the other hand, no hatchability recorded in eggs when exposed to the three tested concentrations of mancozeb. Hatchability rate in eggs when exposed to LC_{25} urea was 77.6%, while it decreased to 27.8% by exposure to LC_{90}. In addition, no hatchability observed when eggs exposed to mixed nitrate.

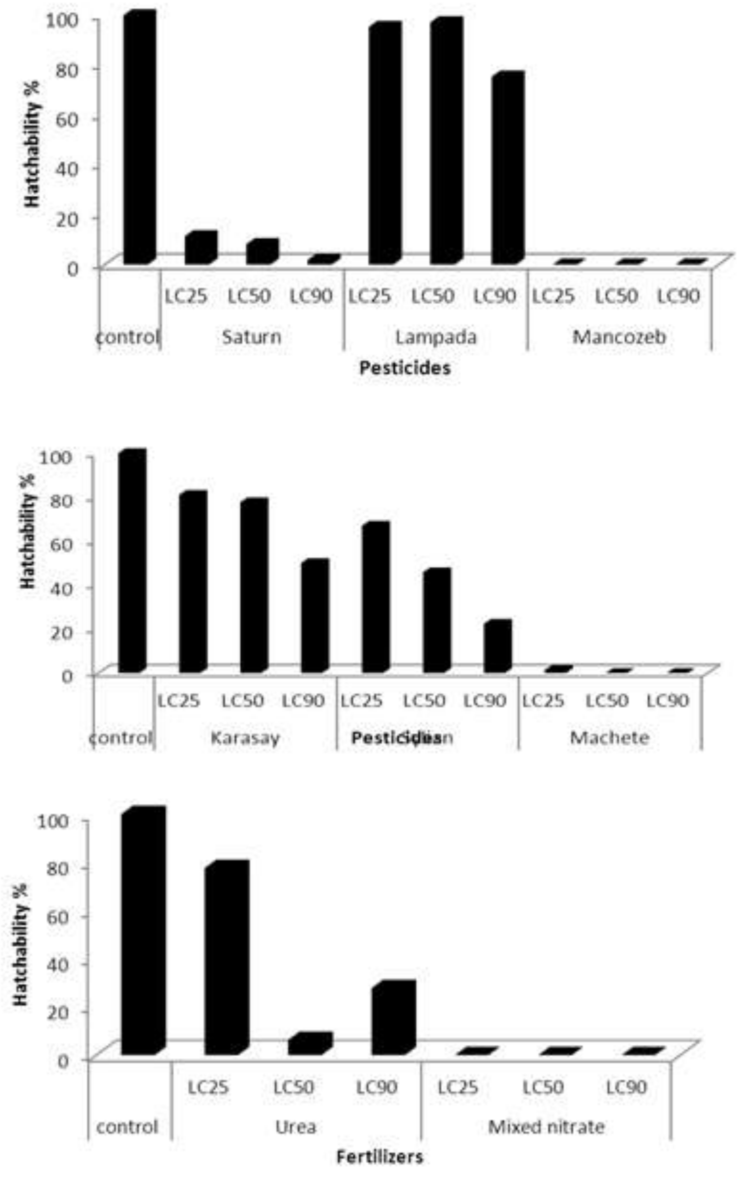

Fig. 2: Impact of pesticides and fertilizers on hatchability of *B. alexandrina* eggs.

Furthermore, the current data (Fig. 3) indicated that more than moderate death rates of *S. mansoni* miracidia were started after minutes of exposure to LC_5 from lampada, sylian, saturn, karasay, mancozeb and machette and the fertilizers urea and mixed nitrate. Thus, after 1 h for LC_5 miracidia death rates were 60%, 51%, 40%, 65%, 20%, 70%, 11%, 47%, respectively, compared to 3% for control group. Moreover, after two hours increased the death rates to 80%, 68%, 65%, 78%, 35%, 87%, 61% and 91%, respectively, compared to 6% for control specimens.

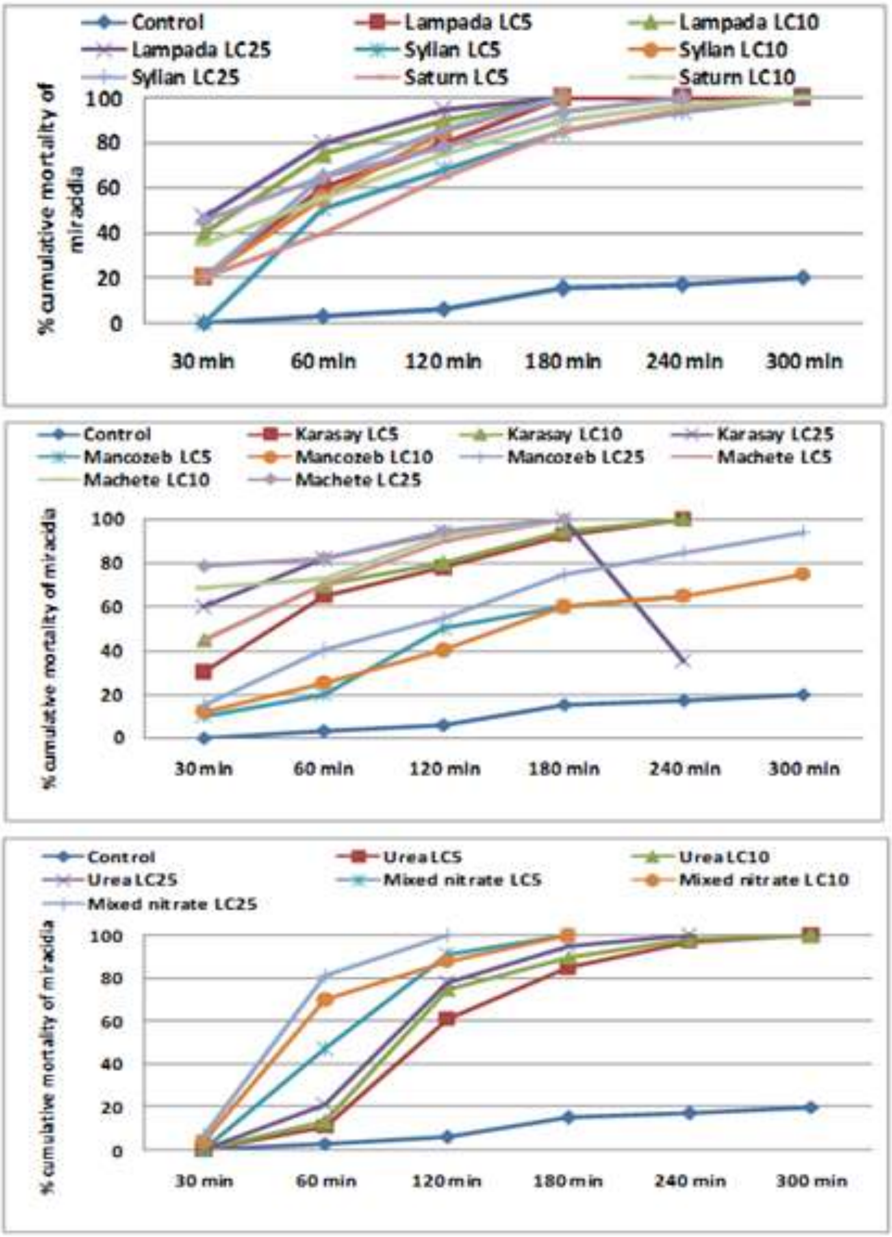

Fig. 3: Effect of pesticides and fertilizers on free living larval stages *of Schistosoma mansoni* miracidi

Moreover, the effect of previous tested materials on cercariae data in Fig (4) indicated that after 30 minutes of cercarial exposure to LC_5 of lampada, sylian, saturn, karasay, mancozeb, machette and urea and mixed nitrate, mortality rates were 15%, 15%, 20%, 25%, 15%, 35%, 15% and 15%, respectively. Elongation of the exposure period to 120 minutes highly raised the mortality rate at the same concentration to be 75%, 75%, 75%, 85%, 70%, 80%, 67% and 66%, respectively. In the meantime, increasing the concentration to LC_{25} lead to increase mortality rates and after 240 minutes of exposure, no cercariae survived.

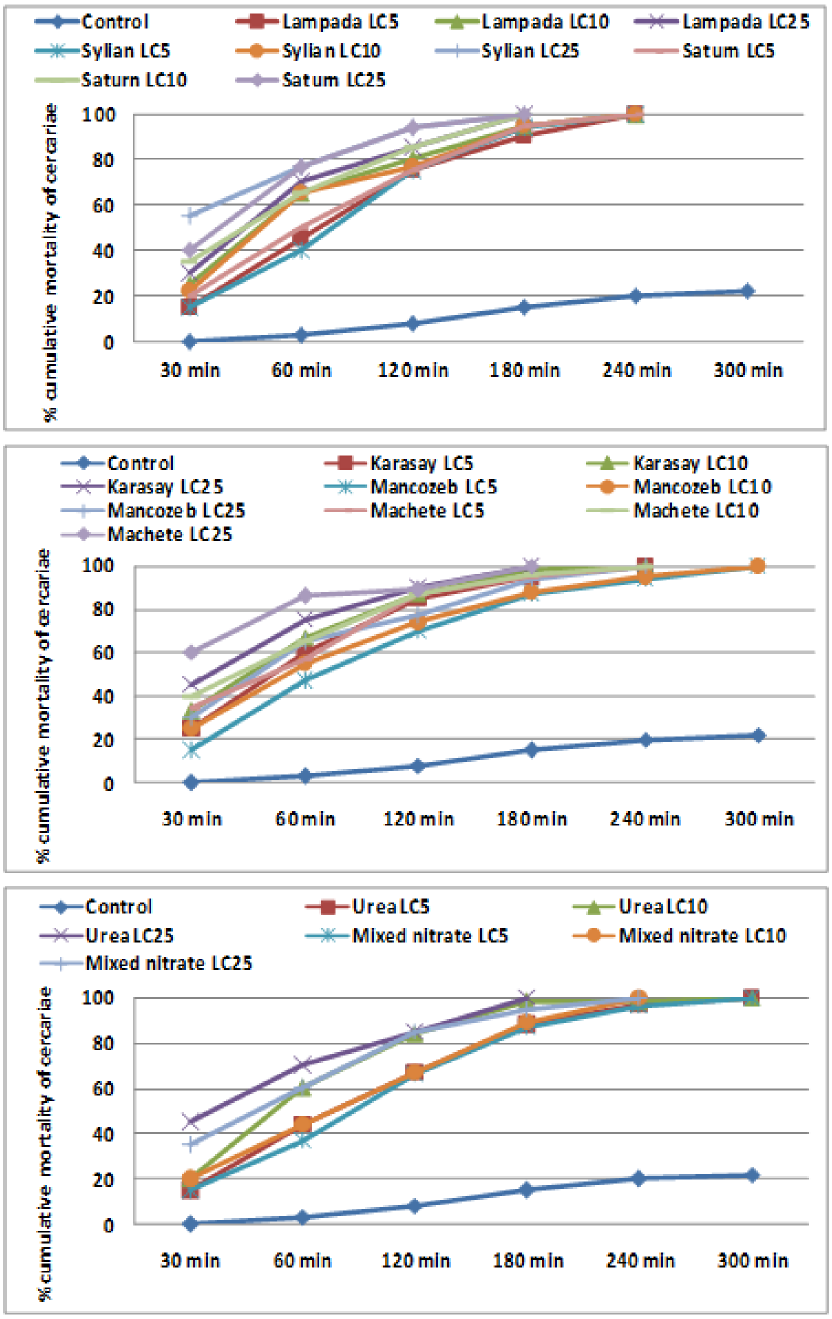

Fig. 4: Effect of pesticides and fertilizers on free living larval stages of *Schistosoma mansoni* cercariae.

DISCUSSION

Irrigated rice cultivation and fish aquacultures contribute to the spread of schistosomiasis as they offer favorable ecological conditions for their intermediate snail hosts and in same time offer closely linked human water contacts (Huang & Manderson, 1992; Bethony *et al.*, 2004 and Steinmann *et al.*, 2006). The present study revealed that *B. alexandrina* and *B. truncatus* are dominant in all watercourses next to non-governmental rice farms and fish aquacultures where *B truncatus* snails showed limited distribution than *B. alexandrina* .The patterns of schistosome snail intermediate hosts distribution and their prevalence of infection are among the measurable indicators that reflect the magnitude of transmission (Sayed *et al.*, 2004). Present results indicated high magnitude of transmission as naturally infected *Biomphalaria* was observed in Kirodrain drains with a percentage 13.3%. in rice farms and El-Tashween with the percentage of 25% for fish aquacultures .While naturally infected *B. truncatus* were found in Menyet Al-Morshed for rice farms and Kotshenar drain for fish aquacultures recorded transmission percentage of 20% and 12.5%, respectively. The present natural infection percentages were higher than that observed by El-Khayat *et al.* (2005) 1.1% among field *B. alexandrina*. Haristone (1973) commented that low percentage of natural infection is the rule (1-2%), however high percentage (3.3-7.5) may be recorded in summer period suggestion that the proportion of snails infected with schistosome at any time depends upon the complex interaction of different factors. This is confirmed by Barboza *et al.* (2012) who concluded that the irrigation in rice cultivate have an effect on the intensity of human infection by *S. mansoni,* and there was a correlation between the levels of *S. mansoni* infection and the time of activities in water sources. Also, Rawi *et al*. (1993) indicated that the introduction of irrigation had increased the exposure to possible transmission of schistosomiasis, directly, by work on irrigated rice-fields.

The present results of physicochemical parameters revealed that all Cu, Na, K & Ca in watercourses of rice farms and Cd, Cu, K and most Na levels in fish aquacultures exceeded the levels of concern that determined by "National Recommended Water Quality Criteria", representing risks on health. Also, they represented negative impacts on the sensitive schistosomiasis free larval stages and consequently transmission. Toxic chemicals and trace metals have a relatively consistent negative effect on intestinal helminthes (Lafferty, 1997). Water pollutants may be kills the sensitive free-living stages of the parasite without adversely affect the intermediate host of that parasite (Evans, 1982). For example, trace metals in sewage sludge reduced survival of free-living cercariae and miracidia, leading to a lower prevalence in the survived snails (Siddall and Des Clers, 1994). Lone *et al*. (2008) reported that the presence of any metal may vary from site to site, depending upon the source of individual pollutant. In the present study Borollos Lake characterized by active *S. mansoni* transmitting sites, high level of certain metals, slightly higher conductivity than canals, high infestation. So, the present results are confirmed by Abdel-Shafy and Aly (2002) who mentioned that all of these characteristics were highly suggested to be as a result of direct discharge of polluted water stream. The Comparative study between governmental and non-governmental farms showed that non-governmental drains were characterized by the most infestation with *B. alexandrina* and *B. truncates* and highest densities of naturally infected snails and revealed that *B. alexandrina* and *B. truncatus* were only observed in non-governmental farms. Comparison of physicochemical parameters the level of water quality in governmental and non-governmental field cleared that pH and K have significantly different values between governmental and non-governmental fish aquaculture. Also, pH and dissolved oxygen were significantly different between watercourses next to governmental and non-governmental rice farms. Dissolved oxygen, Pb, Cu, K, Ca & Na. All Cu, K and Ca in sites next to governmental and non-governmental fish farms exceeded than the water quality standards in governmental and non-governmental agricultural farms. Pesticides may play an important role in the disappearance of schistosome snail intermediate hosts and consequently hindering schistosomiasis transmission Abdel–Kader &Sharaf El-Din (1986) and Ragab and Bakry (2006). This agreed with the present study results which showed that pesticides had strong molluscicidal properties that LC_{50} & LC_{90} of lampada and karasay were 0.8 & 1.0 and 0.7 & 1.1 ppm, respectively but they still less potent than the most applied molluscicide, bayluscide (LC_{50} & LC_{90} are 0.17 & 0.21 ppm). This is parallel with Esmail (2009) who revealed that bayluscide is more toxic to snails than the tested pesticides. Also, the results showed that pesticides were far toxic than fertilizers (LC_{50} & LC_{90} were 2754 & 4092 ppm for mixed nitrate and 9636 & 15937 ppm for Urea). This is confirmed by Ragab and Shokry (2006) who concluded that molluscicidal efficacy of three fertilizers; ammonium nitrate, potassium sulphate and urea had weak efficacy against *B. alexandrina* and their LC_{50} were 470, 1900 and 2200 ppm, respectively. Also, El-Deeb *et al.* (2017) cleared that LC_{50} values were 505.7, 1600 and 9500 ppm for balanced, high phosphorus and high nitrogen fertilizers, respectively. In the same consequence, many studies in Egypt concerned to test many agricultural pesticides and fertilizers for their molluscicidal, ovicidal and larvicidal properties. Abdel Kader and Sharaf El-Din (2005) found that the pesticides Carbofuran and Profenophos usually used in cotton farming and rice farming may be the reason for the disappearance of snail vector from the neighboring water bodies. These findings agree with the present results explored that machete was the most potent against snail eggs as LC_{25}, LC_{50} and LC_{90} concentrations were5.10, 14.80 & 33.20 ppm which killed 94.2 - 100% eggs, with hatchability failure (0 - 4.3%).

This is confirmed by, Ibrahim *et al*. (1992) who reported that exposure of *B. alexandrina* to sublethal concentrations (0.125, 0.25 and 0.05 ppm) of the organ phosphorus insecticide, chlorpyrifos (Dursban), induced a reduction in egg production and egg hatchability, while Hasheesh *et al*. (2011) reported that the high concentrations of the three tested agents Difenoconazole (Score), the plants *Asparagus densiflours* and *Oreopanax guatemalensis* caused a remarkable reduction in hatchability percent of *B. alexandrina* eggs than the low concentrations.

Exposure of *S. mansoni* miracidia and cercariae to the examined pesticides and fertilizers showed close mortality rates that increased by time and concentration suggesting the high sensitivity of these larvae to all the examined chemicals and cercariae were more sensitive than miracidia. Also, El-Emam *et al.* (2008) showed that 0.12 ppm of Match pesticide killed miracidia after 120 minutes while killed cercariae after 90 minutes. Hasheesh and Mohamed (2011) evaluate the larvicidal activity of two pesticides (Chlorpyrifos and Profenophos) and observed that the mortality rates of miracidia and cercariae were elevated gradually by increasing the concentration of the tested pesticides.

CONCLUSION

It is concluded that introduction of such pesticides and fertilizers to snails' habitats through plant pests control, especially the low concentrations of the pesticide mancozeb and mixed nitrate, inhibited the hatchability of *B. alexandrina* eggs, also suppressed vitality of *S. mansoni* larval stages within a short exposure period. As well, they disturbed the compatibility of *B. alexandrina* snails to *S. mansoni*.

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