

Full Length Research Paper

# Toxicity bioassay and effects of sub-lethal exposure of malathion on biochemical composition and haematological parameters of *Clarias gariepinus*

Zubair Ahmad

Department of Zoology, College of Science, P. O. Box 2455, King Saud University, Riyadh 11451, Kingdom of Saudi Arabia. Email: [zahmed@ksu.edu.sa](mailto:zahmed@ksu.edu.sa) or [zubair.ahmad.ah@gmail.com](mailto:zubair.ahmad.ah@gmail.com).

Accepted 13 March, 2020

*Clarias gariepinus* were exposed to different concentrations of malathion to determine the 96 h LC<sub>50</sub> value and its sub-lethal effects on haematological parameters and biochemical composition were also investigated. The 96 h LC<sub>50</sub> value concluded was 8.22 mg/L. Specimens of *C. gariepinus* were exposed to sub-lethal concentrations (0.5, 1.0 and 2.0 mg/L) of pesticide for 4 weeks, which revealed that the pesticide had an adverse effects on various blood parameters. Red blood cell (RBC) and white blood cell (WBC) counts, hemoglobin (Hb) concentration and haematocrit (Ht) values decreased after the exposure of malathion. Plasma glucose level was elevated as plasma protein decreased. Liver and muscle glycogen also decreased in the fish exposed to Malathion. Alanine amino transferase (ALT), glutamate oxaloacetate transaminase (GOT) and glutamate pyruvic transaminase (GPT) activities increased in the fish exposed to malathion. Magnesium and calcium ions were also affected, but the effects were insignificant.

**Key words:** Malathion, bioassay, sub-lethal exposure, *Clarias gariepinus*, biochemical and haematological changes.

## INTRODUCTION

Centuries ago in most parts of the world, pesticides are used to improve crop production by eradicating unwanted insects and human health by controlling undesirable plants, animals as well as disease vectors (Prakasam et al., 2001). Two billion kilograms of pesticides are applied annually to forests, gardens, homes and agricultural lands in United States of America alone (Aspelin and Grube, 1999). Among these pesticides are organophosphorus (OP) compounds commonly used as insecticides. An organophosphorus insecticide, malathion (O-dimethyl S-[1,2-di-(ethoxycarbonyl)ethyl] phosphorodithioate) is commonly used in agriculture and houses to control the variety of insects including aphids, beetles, pill bugs and scales.

Non-target animals including fish are greatly affected by the indiscriminate use of these pesticides. Fish appear to possess the same biochemical pathways to deal with the toxic effects of endogenous and exogenous agents as mammalian species does (Lackner, 1998). Since the fish constitute an important link in food chain and their contamination by pesticides imbalance the aquatic system hence, it is important to examine the toxic effects of pesticides on them.

The haematological parameters like hemoglobin, haematocrit, blood cell counts, glycemia and ion concentrations can be used to find physiological response of contaminated environment (Dethloff et al., 2001). Therefore, when a clinical diagnosis of fish physiology is applied to determine the sub-chronic effects of pollutants, the blood parameters are often measured (Venkataramana et al., 2006). The activities of some enzymes like alanine amino transferase (ALT), glutamate oxaloacetate transaminase (GOT) and glutamate pyruvic transaminase (GPT) also indicate the impacts of pollut-

**Abbreviations:** RBC, Red blood cell; WBC, white blood cell; Hb, hemoglobin; Ht, haematocrit; ALT, alanine amino transferase; GOT, glutamate oxaloacetate transaminase; GPT, glutamate pyruvic transaminase

**Table 1.** Number of dead specimens of *C. gariepinus* and their percentage of mortality (in parentheses) in different concentrations of Malathion at different time intervals.

Concentration (mg/L)	Time (h)			
	24	48	72	96
Control (0.0)	-	-	-	-
7.0	-	-	-	3 (10.00)
7.5	-	-	3 (10.00)	7 (23.33)
8.0	-	3 (10.00)	6 (20.00)	12 (40.00)
8.5	1 (3.33)	3 (10.00)	8 (26.66)	17(56.67)
9.0	3 (10.00)	5 (16.66)	14 (46.66)	22 (73.33)
9.5	5 (16.66)	15 (49.99)	22 (73.33)	27 (90.00)

ants on fish (Bucher and Hofer, 1990). These enzymes are normally found within the cells of liver, heart, gills and kidneys (Shalaby, 2009) but their increase in plasma indicates the tissue injury or organ dysfunction (Wells et al., 1996). However, effects of different pollutants on the biochemical and haematological parameters of fish have been documented (Bucher and Hofer, 1990; Al-Attar, 2005; Ogueji and Auta, 2007; Shalaby, 2009; Abalaka et al., 2011; Al-Kahem Al-Balawi et al., 2011).

*Clarias gariepinus* is an economically important freshwater fish, native to Africa and has been introduced all over the world including Saudi Arabia and form a substantial part of freshwater fishery. In the present study, an attempt was made to investigate the toxicity of malathion to this fish. The mortality of fish and changes in haematological parameters (hemoglobin concentration, cell counts and haematocrit values), biochemical changes (glucose, glycogen and protein content) and enzymes' (ALT, GOT and GPT) activities were monitored after lethal and sub-lethal exposure of this pesticide.

## MATERIALS AND METHODS

Healthy and active specimens of *C. gariepinus* were procured from a fish farm located at Mozamiah, west of Riyadh. The length and weight of fishes ranged from 12 to 14 cm and 55 to 60 g, respectively. The fishes were kept in glass aquaria (160 × 55 × 60 cm) for two weeks to get acclimatized to laboratory conditions. During this period, the commercial fish food were fed twice daily to satiety. Medium of aquaria renewed daily. The water used was analyzed weekly for temperature, dissolved oxygen, hardness and pH, which were recorded as 23.5 ± 1.5°C, 7.5 ± 0.4 mg/L, 230.5 ± 4.5 mg/L as CaCO<sub>3</sub> and 7.8 ± 0.5, respectively. After two weeks of acclimatization, ten fishes were transferred in each aquarium (55 × 30 × 35 cm) containing 30 L of water. Different concentrations (7.0, 7.5, 8.0, 8.5, 9.0 and 9.5 mg/L) of malathion were prepared by adding required volume from the stock solution prepared by diluting the original formulation.

The malathion (MW: 330.4, CAS number: 121-75-5) with 57% active ingredient was obtained from Delta Company, Riyadh. A control set was run with same volume of water and same number of fish. The experiment was run in triplicates. The water was aerated with mechanical pump and feeding was stopped. Dead fishes were removed immediately and their numbers registered. The medium of aquaria was renewed daily. The 96 h LC<sub>50</sub> was computed from a

graph prepared by the method described by Finney (1971). The fishes were exposed for four weeks in triplicates to three different sub-lethal concentrations (0.5, 1.0 and 2.0 mg/L) selected considering the LC<sub>50</sub> value, some blood and biochemical parameters of these exposed specimens were analyzed. A control set was also run for the same time and with the same number of fish but without Malathion. Three fishes from each concentration (one fish from every replicate) were removed after every week during whole experimental period. Blood samples were obtained in heparinized vials by cutting the caudal peduncle; samples of clotted blood were discarded. In case of insufficient quantity, the blood of two or more fishes was pooled.

Hemoglobin was estimated by the cyano-methemoglobin method (Blaxhall and Daisley, 1973). Haematocrit values were determined by using a micro-haematocrit centrifuge. The red blood cell (RBC) and white blood cell (WBC) count was made using Neubauer haemocytometer after diluting the blood with Dace's solution and Turk's solution, respectively. For biochemical analysis, blood was centrifuged at 6000 rpm for 10 min at 4°C and the collected plasma was stored at -20°C till analyzed. Glucose, total protein, calcium (Ca), magnesium (Mg), GOT, GPT and ALT were analyzed using their respective kits (BIOMERIEUX, FRANCE). For statistical analysis, one way analysis of variance (ANOVA) was applied to test the significance of difference among the control and treated values. P values less than 0.05 were considered statistically significant.

## RESULTS

Table 1 shows the mortality of fish as a function of Malathion concentrations. The 96 h LC<sub>50</sub> value for *C. gariepinus* computed from the graph (Figure 1) constructed between log<sub>10</sub> concentrations (X axis) and probit of kill (Y axis) was expressed as 8.22 mg/L. The present findings indicate that in the *C. gariepinus* sub-lethal chronic exposure to malathion altered various blood parameters. The fish exposed to different concentrations of malathion manifested decrease in the erythrocyte and leucocyte counts, hemoglobin concentration and haematocrit values as compared to the control fish (Table 2). A slight change in the value of different indices (MCV, MCH and MCHC) was noticed in *C. gariepinus* after malathion exposure. Significant hyperglycemia and hypo-proteinaemia was evident in the fish exposed to different levels of malathion (Table 3). These changes were more pronounced in the higher

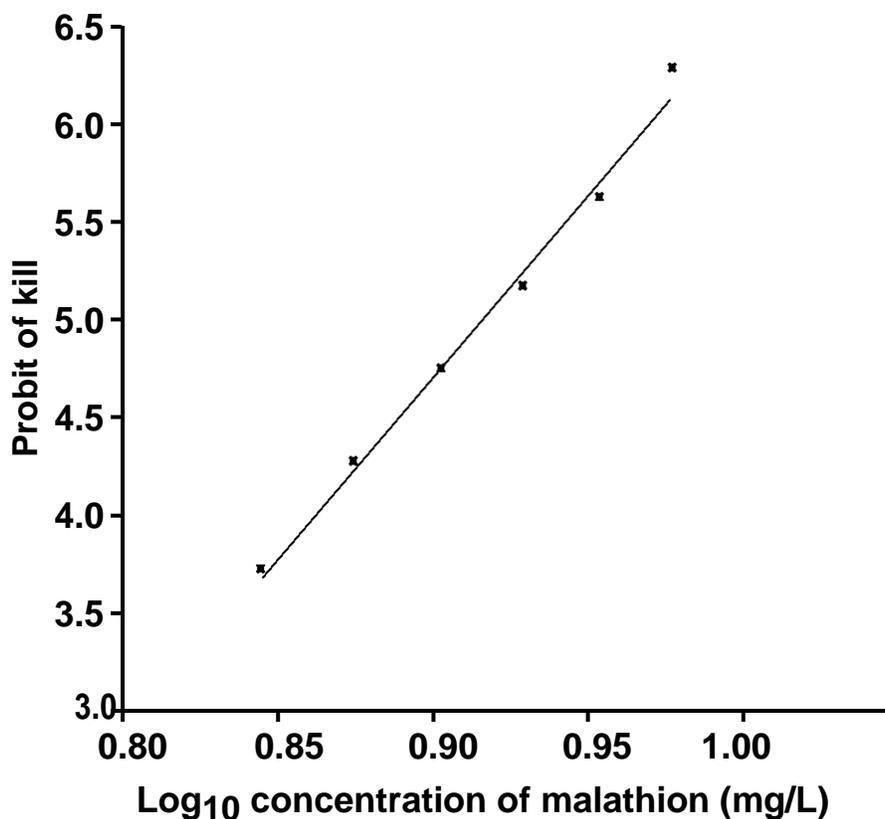


Figure. 1. Graph showing the relationship of probit of kill with log<sub>10</sub> concentration of malathion used to deduce the LC<sub>50</sub> of malathion used to deduce the LC<sub>50</sub>.

50

doses and in the last period of exposure (Table 3). Reduction in the concentration of Ca ions was registered in the fish exposed to high dose of malathion and in the last period of exposure whereas Mg ions remain unchanged.

It is quite apparent from the present investigation that malathion exposure to *C. gariepinus* had markedly elevated the PALT activity (Table 3) in all concentrations tested especially in the last period of exposure. The data embodied in (Table 3) also revealed that the malathion exposure had significantly ( $P < 0.05$ ) enhanced the activity of PGOT and PGPT enzymes after 2 weeks at higher doses (1.0, 2.0 mg/L) and after 4 weeks in all exposed groups.

## DISCUSSION

The LC<sub>50</sub> value (8.22 mg/L) recorded in the present study for *C. gariepinus* is less than the values (9.14 mg/L for *Ptychocheilichthys lucius*, 11700 µg/L for black bullhead, 11.8 mg/L for *Heteropneustes fossilis*, 15.3 mg/L for *Gila elegans* and 17.0 mg/L for *Ictalurus furcatus*) documented by Durkin (2008) and Faria et al. (2010) for various fish species. In contrast to the aforementioned

values, Pathiratne and George (1998) reported a lower 96 h LC<sub>50</sub> value (2.2 ppm) for *Oreochromis niloticus*. Newhart (2006) tabulated the LC<sub>50</sub> values of malathion for different species of fish which ranges from 0.06 to 7620 µg/L. Malathion was found to be highly toxic to fry of *Labeo rohita* (LC<sub>50</sub> value 9 µL, Patil and David, 2008); *Opheocephalus punctatus* (LC<sub>50</sub> 16 µg/L, Pugazhvandan et al., 2009); walleye (LC<sub>50</sub> 64 ppb), brown trout (LC<sub>50</sub> 101 ppb) and cutthroat trout (LC<sub>50</sub> 280 ppb) and moderately toxic to minnows (LC<sub>50</sub> 8.6 ppm) and murrels (LC<sub>50</sub> 5.93 ppm) as summarized by Durkin (2008). The difference in the toxic potential of the pesticides may be attributed mainly to the susceptibility of the test animals and factors like pH and hardness of water.

The disparity in the toxic potential of malathion can also be related to the differences in susceptibility and tolerance related to its accumulation, biotransformation and excretion. Discrepancies in metabolic pathways among species may result in varied patterns of biotransformation, leading to more or less toxic metabolites (Johnsson and Toledo, 1993). The magnitude of toxic effects of pesticides also depends on length and weight, corporal surface/body weight ratio and breathing rate (Murty, 1986). Oh et al. (1991) reported three factors causing selective toxicity of pesticides for various fish

**Table 2.** Effects of Malathion exposure on hematological parameters of *Clarias gariepinus*.

Parameter	Concentration (mg/L)	Exposure time (week)			
		1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>
Erythrocytes (Cellx106/mm3)	Control (0.0)	1.66 ± 0.05	1.64 ± 0.061	1.60 ± 0.054	1.65 ± 0.046
	0.5	1.61±0.05	1.60 ± 0.042	1.60 ± 0.042	1.61 ± 0.054
	1.0	1.58 ± 0.08	1.58 ± 0.053	1.46 ± 0.058*	1.43 ± 0.061*
	2.0	1.55 ± 0.06*	1.53 ± 0.061*	1.52 ± 0.045*	1.41 ± 0.046*
Leucocytes (Cellx103/mm3)	Control (0.0)	36.51 ± 0.51	37.23 ± 0.65	37.21 ± 0.71	38.54 ± 0.54
	0.5	33.02 ± 0.52	34.56 ± 0.52	32.65 ± 0.65	35.01 ± 0.62
	1.0	33.56 ± 0.44	31.89 ± 0.57*	31.12 ± 0.50*	30.01 ± 0.42*
	2.0	31.45 ± 0.61*	31.01 ± 0.43*	30.25 ± 0.59*	28.54 ± 0.24*
Haematocrit (%)	Control (0.0)	33.06 ± 0.72	34.76 ± 0.50	33.44 ± 1.02	34.21 ± 0.62
	0.5	32.54 ± 0.51	33.21 ± 0.82	32.23 ± 0.56	33.80 ± 0.96
	1.0	32.65 ± 0.92	32.35 ± 1.10	32.21 ± 1.05	31.65 ± 1.10
	2.0	31.45 ± 0.94*	31.85 ± 0.96*	30.42 ± 1.20*	30.25 ± 1.09*
Hemoglobin (g/dl)	Control (0.0)	5.65 ± 0.09	6.01 ± 0.12	5.95 ± 0.10	6.12 ± 0.61
	0.5	5.11 ± 0.11	5.15 ± 0.13	5.35 ± 0.10	5.56 ± 0.29
	1.0	4.54 ± 0.14*	4.64 ± 0.13*	5.66 ± 0.09	4.54 ± 0.11*
	2.0	4.24 ± 0.09*	4.29 ± 0.08*	4.24 ± 0.12*	4.14 ± 0.12*
MCV (fl/cell)	Control (0.0)	199.92 ± 3.75	211.96 ± 4.21	209.00 ± 3.58	207.33 ± 4.12
	0.5	202.54 ± 3.44	207.56 ± 4.21	201.44 ± 4.23	209.94 ± 5.56
	1.0	206.65 ± 4.35	204.74 ± 3.56	206.47 ± 5.21	221.33 ± 5.25*
	2.0	202.90 ± 4.33	208.17 ± 4.25	200.13 ± 5.65	209.60 ± 5.95
MCH (Pg/cell)	Control (0.0)	34.04 ± 1.75	36.65 ± 1.65	37.19 ± 2.11	37.09 ± 1.75
	0.5	31.74 ± 1.64	32.29 ± 2.33	33.44 ± 1.45	34.53 ± 2.25
	1.0	28.73 ± 2.55	29.36 ± 2.45	29.87 ± 1.75	31.75 ± 2.35*
	2.0	27.35 ± 2.32*	28.04 ± 1.45*	27.89 ± 2.45*	27.42 ± 1.46*
MCHC (%)	Control (0.0)	17.09 ± 1.35	17.29 ± 1.46	17.79 ± 1.25	17.89 ± 1.01
	0.5	15.70 ± 0.65	15.51 ± 0.95	16.60 ± 1.15	16.45 ± 0.98
	1.0	13.90 ± 1.15	14.34 ± 1.26	14.47 ± 0.85	14.34 ± 0.75
	2.0	13.48 ± 1.15	13.47 ± 1.45	13.94 ± 1.65*	13.80 ± 1.45*

\*Significant difference with control (P<0.05). Values are mean ± standard error.

species which are varied inhibition of acetylcholinesterase, detoxification and absorption. In general, the toxicity varied with respect to species, size of fish and duration of exposure (Oh et al., 1991; Dutta et al., 1995). Blood parameters, generally, of fish are considered as suitable tool for evaluating the effects of chemicals. Past investigators have also identified changes in several haematological parameters as indicators of pollutants exposure specially metals (Cyriac et al., 1989).

Reduction in different blood parameters might be due to malfunctioning of the haematopoietic system caused by Malathion exposure. Similar to the present results, a

decrease in the number of RBC, hemoglobin and haematocrit values of diazinon (an organophosphate pesticide) exposed fish was reported by Banaee et al. (2008, 2011) and related it to destruction of cells and/or decrease in size of cells due to the adverse effects of pesticide. Zaki et al. (2009) reported that RBC count, hemoglobin concentration and PVC values were dwindled in the fish exposed to malathion. Adeyemo (2007) reported decreased hemoglobin, RBC count and haematocrit values in *C. gariepinus* exposed to lead nitrate. Generally, toxicants exposure exerts an adverse effect on the haematopoietic organs which in turn alters

**Table 3.** Effects of malathion exposure on biochemical composition of *Clarias gariepinus*.

Parameter	Concentration (mg/L)	Exposure time (week)			
		1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>
Total Protein (g/dl)	Control (0.0)	28.75 ± 1.78	29.35 ± 1.87	29.65 ± 2.09	28.85 ± 2.05
	0.5	28.65 ± 1.86	29.65 ± 1.68	27.25 ± 1.22	26.56 ± 1.96
	1.0	27.98 ± 1.98	27.65 ± 1.02	27.05 ± 1.95	26.85 ± 1.75
	2.0	28.05 ± 1.88	27.85 ± 1.75	26.25 ± 1.86	22.60 ± 1.85*
Glucose (mg/100ml)	Control (0.0)	45.25 ± 7.25	48.35 ± 6.68	46.52 ± 6.88	45.95 ± 6.88
	0.5	55.25 ± 7.55	58.25 ± 7.25	65.55 ± 5.85*	60.25 ± 7.88*
	1.0	65.25 ± 8.68*	64.65 ± 8.25*	78.54 ± 6.75*	70.25 ± 7.52*
	2.0	70.25 ± 7.56*	72.25 ± 7.54*	90.25 ± 8.25*	95.25 ± 6.25*
Liver glycogen (mg/g)	Control (0.0)	9.12 ± 0.12	8.92 ± 0.21	8.90 ± 0.17	8.91 ± 0.18
	0.5	8.76 ± 0.17	8.25 ± 0.18	8.45 ± 0.16	8.44 ± 0.18
	1.0	7.25 ± 0.16*	7.25 ± 0.19*	7.25 ± 0.16*	7.35 ± 0.18*
	2.0	7.36 ± 0.15*	7.32 ± 0.17*	7.35 ± 0.15*	7.25 ± 0.18*
Muscle glycogen (mg/g)	Control (0.0)	3.45 ± 0.08	3.35 ± 0.06	3.35 ± 0.06	3.31 ± 0.05
	0.5	3.25 ± 0.06	3.25 ± 0.06	3.10 ± 0.07	2.94 ± 0.05
	1.0	2.15 ± 0.06*	2.12 ± 0.05*	2.10 ± 0.05*	2.00 ± 0.05*
	2.0	2.05 ± 0.05*	2.06 ± 0.05*	2.06 ± 0.06*	2.15 ± 0.05*
Ca (mg/dl)	Control (0.0)	180.45 ± 12.3	190.25 ± 10.2	200.45 ± 11.2	202.45 ± 10.6
	0.5	175.35 ± 10.1	165.25 ± 11.2	180.95 ± 12.3	190.65 ± 11.4
	1.0	165.25 ± 11.2	162.45 ± 12.3	155.25 ± 10.6*	160.25 ± 12.3
	2.0	155.65 ± 13.1*	155.35 ± 09.6*	150.65 ± 08.9*	143.35 ± 11.8*
Mg (mg/dl)	Control (0.0)	38.25 ± 3.12	36.03 ± 2.15	35.32 ± 4.25	36.45 ± 4.56
	0.5	42.45 ± 2.25	35.24 ± 3.25	36.24 ± 5.25	37.54 ± 4.26
	1.0	41.24 ± 5.31	37.45 ± 5.21	37.45 ± 5.23	35.24 ± 5.35
	2.0	40.23 ± 5.51	36.56 ± 3.45	36.45 ± 4.24	3.25 ± 4.45
PALT (IU/l)	Control (0.0)	50.43 ± 3.56	53.25 ± 2.65	47.53 ± 3.21	51.23 ± 3.65
	0.5	55.32 ± 4.21	62.21 ± 3.23	60.23 ± 3.54	63.12 ± 3.68*
	1.0	58.56 ± 3.21	63.21 ± 3.56*	63.21 ± 4.05*	65.32 ± 4.36*
	2.0	62.35 ± 4.03*	65.32 ± 4.12*	67.25 ± 3.87*	70.23 ± 4.06*
PGOT (IU/l)	Control (0.0)	80.25 ± 15.2	85.32 ± 13.5	85.26 ± 12.4	80.65 ± 10.5
	0.5	95.25 ± 13.3	101.25 ± 16.2	108.23 ± 11.2*	111.35 ± 11.3*
	1.0	97.25 ± 16.2*	105.35 ± 14.3*	110.45 ± 10.5*	115.65 ± 10.2*
	2.0	101.25 ± 14.2*	120.25 ± 16.2*	125.35 ± 14.2*	135.12 ± 11.2*
PGPT (IU/l)	Control (0.0)	65.12 ± 5.66	68.22 ± 8.11	68.21 ± 6.84	70.21 ± 7.66
	0.5	75.21 ± 8.55	75.44 ± 6.88	82.23 ± 8.25	80.25 ± 8.25
	1.0	85.25 ± 11.3*	86.25 ± 10.3*	83.25 ± 11.2*	92.25 ± 6.21*
	2.0	99.25 ± 11.2*	92.35 ± 12.1*	109.25 ± 11.1*	114.23 ± 11.3*

\*Significant difference with control (P<0.05). Values are mean ± standard error.

blood parameters. Changes in the leukocyte system manifest in the form of leukocytosis with heterophilia and lymphopenia, which are characteristics of leukocyte

response in animals exhibiting stress. Al-Kahem (1995) reported reduced WBC count in the fish exposed to chromium and acclaimed it to be a consequence of

significant decline in the number of lymphocytes and thrombocytes. Reduction in the number of lymphocytes count in the fish, *Oreochromis niloticus*, exposed to trichlorfon was attributed to fall in the delivery of these cells to the circulation because of reduced production or alternatively an increased rate of removal from circulation and subsequent rapid destruction of cells. Leukocyte count diminished in tilapia exposed to phosalone (Jaffar and Rani, 2009).

The blood cell indices like mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) seem to be changes that are more sensitive and can cause reversible changes in the homeostatic system of fish. Fluctuations in these indices correspond with values of RBC count, hemoglobin concentration and packed cell volume. The values of blood cell indices were enhanced in common carp and other freshwater fish after the exposure of acute toxic level of pesticides (Rao, 2010). The elevated level of glucose expressed in the blood of Malathion exposed fish may be due to the mobilization of glycogen into glucose to meet the increased demand for energy. Glucocorticoids and catecholamine hormones are known to produce hyperglycemia in animals and stress stimuli elicit rapid secretion of these hormones from adrenal tissue of the fish (Pickering, 1981). Such elevation may be due to enhanced gluconeogenesis response of stressed fish in their attempt to satisfy their new energy demands (Winkler et al., 2007).

The hyperglycemic condition in the present study may also be attributed to increased secretion of these hormones which causes glycolysis in the fish exposed to Malathion. The present result agrees with the findings of Abalaka et al. (2011) and Alkahem-Al-Balawi et al. (2011). The pesticide may change the functions of vital organs like liver and kidney, disrupting the homeostatic condition of the body which may alter the concentrations of metals. Similar observations have been reported by Al-Akel et al. (2010) in the carp, *Cyprinus carpio*, after the exposure of dietary copper and support to the present investigation. The reduction in the protein level in the fish exposed to toxicants can be attributed to the cellular destruction or necrosis with subsequent impairment of protein synthesis machineries (Bradbury et al., 1987) or due to pathological alterations in kidney leading to excessive loss of proteins (Salah El-Deen et al., 1996). However, the hypoproteinaemia in the present study may also be ascribed to the aforementioned factors. Omoniyi et al. (2002) and Shalaby (2009) have reported hypoproteinaemia in the fish exposed to pollutants. In contrast to the present findings, a hyperproteinaemia was reported by Al-Attar (2005), Omitoyin (2007) and Abalaka et al. (2011). They were of the opinion that hyperproteinaemia may be the repercussion of water loss in plasma, elevated *de novo* synthesis or relative changes in blood protein mobilization. They also mentioned that such observed hyperproteinaemia may be

indicative of efficient immune response and body physiological reaction to pollutants. An elevated level of ALT activity in fish exposed to malathion and extract of *Porkiabiiglosa* pods was documented by Zaki et al. (2009) and Abalaka et al. (2011), respectively. These authors believe that the increased activity of enzyme in the exposed fish is suggestive of hepatic damages leading to their leakage in circulation (Mousa et al., 2008) and /or increased synthesis of enzyme in liver. Contrary to this, some authors like Okechukwu and Auta (2007) and Hedayati et al. (2010) reported that the ALT activity in the fish exposed to different pollutants was inhibited. This reduction in the enzyme activity was attributed to liver necrosis caused by toxicants and a possible damage to hepatocytes or low sub-lethal doses of toxicants used to expose the fish. SGPT and SGOT enzymes are supposed to be sensitive to any change in the environment. Therefore, the exposure of fish to the pollutants expresses elevated level of these enzymes. Jeney et al. (1991) reported an elevated level of these enzymes (SGOT, SGPT) in the serum of fish exposed to ammonia.

Their conception was that SGPT is highly sensitive to alterations in the environmental condition. Similarly, significantly higher values of glutamate oxaloacetate acid transaminase (GOT) activities were recorded by lemaire et al. (1991) in the fish fed diet without docosahexaenoic acid (DHA). Contrary to this, the activity of GPT did not show any change. They found that hepatic parenchyma develop into generalized massive steatosis, exhibiting necrosis centers with docosahexaenoic acid free diet. Exposure of monocrotophos to *Corydoras punctatus* increased the activity of SGOT and SGPT (Agrahari et al., 2007).

In addition, Palanivelu et al. (2005) suggested that liver is rich in SGOT and SGPT, and damage to it could result in liberation of large quantities of these enzymes into the blood.

Hence, an increase in the activity of these enzymes (PGOT and PGPT) after the pollutants treatment is a sensitive indicator of cellular damage (Palanivelu et al., 2005; Alkahem Al-Balawi et al., 2011). Therefore, higher activities of these enzymes registered in the present investigation may be ascribed to damage caused to liver by malathion.

## Conclusion

Malathion seems to be moderately toxic to *C. gariepinus*. The LC<sub>50</sub> (8.22 mg/L) registered were within the values obtained for other species of fish. The present study enhanced the knowledge of biochemical and haematological alterations in fish due to chronic sub-lethal exposure of Malathion.

The data obtained in the present investigation amply emphasized that malathion had adverse effects on the metabolism of macromolecule and haematopoietic

organs of fish. Therefore, the use of pesticide in the field may be a threat to human, fauna and flora of the environment.

## ACKNOWLEDGEMENT

The King Saud University, Deanship for Scientific Research, Research Center, College of Science, is gratefully acknowledged for supporting the present research work.

## REFERENCES

- Abalaka SE, Esievo KAN, Shoyinka SVO (2011). Evaluation of biochemical changes in *Clarias gariepinus* adults exposed to aqueous and ethanolic extracts of *Parkia biglobosa* pods. *Afr. J. Biotechnol.* 10: 234-240.
- Adeyemo OK (2007). Haematological profile of *Clarias gariepinus* (Burchell, 1822) exposed to lead. *Turkish J. Fish Aquat. Sci.* 7: 163-169.
- Agrahari S, Pandey KC, Gopal K (2007). Biochemical alteration induced by monocrotophos in the blood plasma of fish, *Channa punctatus* (Bloch). *Pest. Biochem. Physiol.* 88: 268-272.
- Al-Akel AS, Alkahem-Al-Balawi HF, Al-Misned F, Mahboob S, Ahmad Z, Suliman EM (2010). Effects of dietary copper exposure on accumulation, growth, and hematological parameters in *Cyprinus carpio*. *Toxicol. Environ. Chem.* 92: 1865-1878.
- Al-Attar AM (2005). Biochemical effects of short-term cadmium exposure on the freshwater fish, *Oreochromis niloticus*. *J. Biol. Sci.* 5: 260-265.
- Alkahem HF (1995). Behavioral responses and changes in some haematological parameters of the cichlid fish, *Oreochromis niloticus*, exposed to trivalent chromium. *J. King Abdul Aziz Univ. Sci.* 7: 5-13.
- Alkahem Al-Balawi HF, Ahmad Z, Al-Akel AS, Al-Misned F, Suliman EM, Al-Ghanim KA (2011). Toxicity bioassay of lead acetate and effects of its sub-lethal exposure on growth, haematological parameters and reproduction in *Clarias gariepinus*. *Afr. J. Biotech.* 10: 11039-11047.
- Aspelin AL, Grube AH (1999). Pesticide industry sales and usage 1996 and 1997 market estimates, EPA 733-R-99-001, Office of pesticide programs, Washington, DC.
- Banaee M, Mirvaghefi AR, Rafei GR, Majazi Amiri B (2008). Effects of sub-lethal diazinon concentrations on blood plasma biochemistry of common carp. *Int. J. Environ. Res.* 2: 189-198.
- Banaee M, Sureda A, Mirvaghefi AR, Ahmadi K (2011). Effects of diazinon on biochemical parameters of blood in rainbow trout (*Oncorhynchus mykiss*). *Pest. Biochem. Physiol.* 99:1-6.
- Blaxhall PC, Daisley KW (1973). Routine haematological methods for use with fish blood. *J. Fish Biol.* 5: 771-781.
- Bradbury SP, Symonic DM, Coats JR, Atchison GJ (1987). Toxicology of fenvalerate and its constituent isomers to the fathead minnow (*Pimephales promelas*) and blue gill (*Lepomis macrochirus*). *Bull. Environ. Cont. Toxicol.* 38: 727-735.
- Bucher F, Hofer R (1990). Effect of domestic wastewater on serum enzyme activities of brown trout (*Salmo trutta*). *Comp. Biochem. Physiol.* 97: 385-390.
- Cyriac PJ, Antony A, Nambison PNK (1989). Hemoglobin and hematocrit values in the fish *Oreochromis mossambicus* (Peters) after short term exposure to copper and mercury. *Bull. Environ. Cont. Toxicol.* 43: 315-320.
- Dethloff GM, Bailey HC, Maier KJ (2001). Effect of dissolved copper on selected haematological, biochemical and immunological parameters of wild rainbow trout (*Oncorhynchus mykiss*). *Archi. Environ. Cont. Toxicol.* 40: 371-380.
- Durkin PR (2008). Malathion; Human Health and ecological risk assessment. Final report submitted to Paul Mistretta, PCR, USDA/Forest Service, Southern region, Atlanta Georgia. SERA TR-052-02-02c, p. 325.
- Dutta HM, Munshi JSD, Dutta GR, Singh NK, Adhikari S, Richmonds CR (1995). Age related differences in the inhibition of brain acetylcholinesterase activity of *Heteropneustes fossilis* (Bloch) by malathion. *Comp. Biochem. Physiol.* 111A: 331-334.
- Faria IR, Palumbo AJ, Fojut TL, Tjeerdema RS (2010). Water quality criteria report for malathion. Phase III: Application of the pesticide water quality criteria methodology. UCDAVIS, 7: p. 64.
- Finney DJ (1971). Probit analysis. S. Chand and Company Ltd. Ram Nagar, Delhi.
- Hedayati A, Safahieh A, Savari A, Marammazi JG (2010). Assessment of aminotransferase enzymes in yellowfin sea bream (*Acanthopagrus latus*) under experimental condition as biomarkers of mercury pollution. *World J. Fish Mar. Sci.* 2:186-192.
- Jaffar Ali HA, Rani VJ (2009). Effect of phosalone on haematological indices in the tilapia, *Oreochromis mossambicus*. *Turk. J. Vet. Anim. Sci.* 33:407-411.
- Jeney G, Nemcsok J, Jeney ZS, Olah J (1991). Acute effect of sublethal ammonia concentrations on common carp (*Cyprinus carpio* L.). II. Effect of ammonia on blood plasma transaminases (GOT, GPT), GDH enzyme activity and ATP value. *Aquaculture*, 104: 149-156.
- Johnsson CM, Toledo MCF (1993). Acute toxicity of endosulfan to the fish *Hyphessobrycon bifasciatus* and *Brachydanio rerio*. *Archiv Environ. Conta. Toxicol.* 24: 151-155.
- Lackner R (1998). Oxidative stress in fish by environmental pollutants. *Ecotoxicol.*, pp. 203-224.
- Lemaire P, Draï P, Mathieu A, Carriere S, Giudicelli J, Lafaurie M (1991). Changes with different diets in plasma enzymes (GOT, GPT, LDH, ALP) and plasma lipids (Cholesterol, triglycerides) of sea- bass (*Dicentrarchus labrax*). *Aquaculture*, 93: 63-75.
- Mousa MMA, El-Ashram AMM, Hamed M (2008). Effects of Neem leaf extract on freshwater fishes and zooplankton community. 8<sup>th</sup> International symposium on tilapia in aquaculture. The Central Laboratory for Aquaculture Research, Cairo, Egypt. Oct. 12-14.
- Murty AS (1986). Toxicity of pesticides to fish. CRC Press Inc Boca Raton, FL. p. 143
- Newhart KL (2006). Environmental fate of malathion. California Environmental protection Agency. p. 20.
- Ogueji EO, Auta J (2007). Investigations of biochemical effects of acute concentrations of Lambda-cyhalothrin on African catfish, *Clarias gariepinus* –Teugels. *J. Fish. Int.* 2: 86-90.
- Oh HS, Lee SK, Kim YH, Roh JK (1991). Mechanism of selective toxicity of diazinon to killifish (*Oryzias latipes*) and loach (*Misgurnus anguillicaudatus*). *Aquat. Toxicol. Risk Assess.* 14: 343-353.
- Okechukwu EO, Auta J (2007). The effects of sublethal doses of Lambda-cyhalothrin on some biochemical characteristic of the African catfish, *Clarias gariepinus*. *J. Biol. Sci.* 7: 1473-1477.
- Omitoyin BO (2007). Plasma biochemistry changes in *Clarias gariepinus* (Buchell, 1822) fed poultry litter. *Asian J. Anim. Sci.* 7: 45-52.
- Omoniyi I, Agbon AO, Sodunke SA (2002). Effects of lethal and sub-lethal concentrations of Tobacco (*Nicotiana tobaccum*) leaf dust extract on weight and haematological changes in *Clarias gariepinus* (Burchell) *J. Appl. Sci. Environ. Manage.* 6: 37-41.
- Palanivelu P, Vijayavel K, Ezhilarasibalasubramanian S, Balasubramanian MP (2005). Influence of insecticidal derivatives (Cartap Hydrochloride) from the marine polychaete on certain enzymes of the freshwater fish *Oreochromis mossambicus*. *J. Environ. Biol.* 26: 191-196.
- Pathiratne A, George SG (1998). Toxicity of malathion to Nile tilapia, *Oreochromis niloticus* and modulation by other environmental contaminants. *Aquat. Toxicol.* 43: 261-271.
- Patil VK, David M (2008). Behaviour and respiratory dysfunction as an index of malathion toxicity in the freshwater fish *Labeo rohita* (Hamilton). *Turk. J. Fish. Aquat. Sci.* 8: 233-237.
- Pickering AD (1981). Stress and compensation in teleostean fishes: Response to social and physical factors. In: Pickering, AD (ed.) Stress and fish. Academic Press, New York, USA. pp. 295-322.
- Prakasam A, Sethupathy S, Lalitha S (2001). Plasma and RBCs antioxidant status in occupational male pesticide sprayers. *Clin. Chem. Acta.* 310: 107-112.
- Pugazhvendan SR, Narendiran NH, Kumaran RG, Kumaran S,

- Alagapan KM (2009). Effect of malathion toxicity in the freshwater fish *Ophoecephalus punctatus*-A histological and histochemical study. *World J. Fish Mar. Sci.* 1: 218-224.
- Rao DS (2010). Carbaryl induced changes in the haematological, serum biochemical and immunological responses of common carp, *Cyprinus carpio*, (L.) with special emphasis on herbal extracts as immunomodulators. Ph. D. Thesis, Andhra University, India. p. 235.
- Salah El-Deen MA, Sharada HI, Abu-El-Ella SM (1996). Some metabolic alteration in grass carp (*Ctenopharyngodon idella*) induced by exposure to cadmium. *J. Egypt. Ger. Soc. Zool.* 21: 441-457.
- Shalaby AME (2009). The opposing effects of ascorbic acid (Vitamin C) on ochratoxin toxicity in Nile tilapia (*Oreochromis niloticus*). <http://www.ag.arizona.edu/ista/ista6web/pdf/209.pdf>. Retrieved: 05-04-09.
- Venkataramana GV, Sandhya Rani PN, Murthy PS (2006). Impact of malathion on the biochemical parameters of gobiid fish, *Glossogobius giurus* (Ham). *J. Environ. Boil.* 27: 119-122.
- Wells RM, McIntyre RH, Morgan AK, Davis PS (1996). Physiological stress responses in big gamefish after exposure: Observations on plasma chemistry and blood factors. *Comp. Biochem. Physiol.* 84: 565-571.
- Winkaler EU, Santosh TRM, Machdo-Neto JG, Martinez CBR (2007). Acute lethal and sub-lethal effects of neem leaf extracts on neotropical freshwater fish, *Prochilodus lineatus*. *Comp. Biochem. Physiol. Part C*, 145: 236-244.
- Zaki MS, Mostafa SO, Nasr S, Noor El-Deen AI, Ata NS, Awad IM (2009). Biochemical, clinicopathological and microbial changes in *Clarias gariepinus* exposed to pesticide malathion and climate changes. *Reports Opinion*, pp. 6-11.