

Effect of Malafos (57 EC) Acute Toxicity on Fresh Water Fish *Oreochromis Niloticus*

Hassan Ali Osman

A.R. Prof. Department of aquaculture and fish breeding, Fisheries and Aquatic Research Center Animal Resources Research Corporation, Ministry of Animal Resources and Fisheries–Khartoum–Sudan.

**Corresponding author:* Hassan Ali Osman, A.R. Prof. Department of aquaculture and fish breeding, Fisheries and Aquatic Research Center Animal Resources Research Corporation, Ministry of Animal Resources and Fisheries–Khartoum–Sudan.

ABSTRACT

The present study designed to investigate the effects of acute toxicity of malafos on behavioral and some haematological parameters in the fresh water fish *Oreochromis niloticus*. The fishes were collected from Fisheries and Aquatic Research Center farm in Khartoum and private farm at Alshagera –Khartoum, fish were exposed to different concentrations of malafos (0.75, 1.00, 1.25, 1.50 and 1.75 mg/l) to determine the 96 h LC_{50} value which was (1.0125 ppm) of Malafos 57 EC for *O.niloticus*. Behavioral abnormalities were recorded in fish exposed to different concentrations levels of malafos. The present result revealed that malafo causes alterations in blood parameters. Red (RBCs), hemoglobin (Hb) and hematocrit (Ht%) show significant decrease while white blood cells (WBCs) show increase in their number.

Keywords: malafos, *Oreochromis niloticus*, behavioral abnormalities, hematological change

INTRODUCTION

Oreochromis niloticus is now one of the most widely distributed exotic fish in the world, second only to common carp, as their introduced range now stretches to nearly every continent and include 90 different countries (Intervet, 2006). It is widespread in the tropics and sub-tropics because they are highly adoptable and highly fecund with high fry availability (Seafood Watch, 2006). One stressor influencing fish health is that imposed by pollution. For centuries pesticides have been used in agriculture to improve food production by eliminating unwanted pests and controlling disease vectors (A. Prakasam and S.Sethupathy, 2001). Among common pesticides, organophosphorus compounds are broadly used in medicine industry and cultivation. (E. Storm et al 2000). Malathion is wide spectrum pesticide in organophosphate family and is extensively used throughout the world, (P.D.Moore et al ,2011). Malathion is used excessively to protect crops which eventually affect the aquatic ecosystem including fishes. Malathion gets oxidized to malaaxon by cytochrome p450 enzymes and it is considered to be more toxic than the main compound, (E.Bonilla et al ; 2008). The primary site of action of malathion is the central

and peripheral nervous systems because they inhibit Acetylcholinesterase, (L.W.H.U. Chandrasekara and A.Pathiratne, 2007). Many reports have indicated that malathion, even at low concentration, harms fish, alterate growth parameters. Hematological properties, swimming ability and depletion of some biochemical parameters. (Huculeci R., et al; 2009). (Ahmad; 2011.) Insecticides may lead to changes in the blood and haematological profile of fish (Banaee et al., 2008; Kavitha and Rao, 2009). Acute toxicity caused by different toxicant on freshwater fish can be evaluated by quantitative parameters like survival and mortality of test fish and sensitivity of different fish species against metal toxicity. (Reda, et al; 2010. Misha A., et al 2012). The behavior study is becoming prominent in toxicity assessment in fish, (hansen JA et al 1999. Rao JV, et al 2005). The fingerlings showed initial disturbed behaviours (Amos B. Kayode et al 2001). Ramesh Halappa and Muniswamy David 2009). The purpose of this study to determine LC_{50} of Malafos 57 EC and to investigate the effects of acute toxicity of malafos on behavioral and some haematological parameters in the fresh water fish *Oreochromis niloticus*.

MATERIALS AND METHODS

Chemical

Malafos 57 EC Insecticide. From Medmac for manufacturing Agriculture Chemicals & Veterinary products- Amman-Jordan. Chemical name : malathion; (O-O-Dimethyl phosphorodi thioate of diethyl mercaptosuccinate). Chemical Family; Organophosphate Insecticide is an organophosphate insecticide for use on a variety of agricultural crops, including fruit, vegetable, nut, palme, field crops and non-agricultural premise use

Fish

120 fish of *Oreochromis niloticus* were collected from Fisheries Aquatic research center farm in Khartoum and private farm at Alshagera –Khartoum. Fish acclimated in the lab. for seven days during which fish fed with commercial food(35%)protein diet. Fish were starved one day before the experiment and throughout the test. The physical and chemical parameters of water were obtained daily using Hanna water kids (Ph7.6±0,4 DO6.1±0.5) daily water temperatures ranged from 29 to 31° C,

Experiment

After acclimation 70 of the collected fishes were divided into seven groups each of 10 fishes with average weight 49 ±4 in glass aquaria(40*40 *100 cm) each filled with 70L of unchlorinated water. Malafos six experimental aquaria two replicates per concentration and control set was run with same number of fish and same volume of water and aquarium, continuously aerated using air pumps.

Table1. Determination of Half lethal concentration (LC50)/96h of malafos 57EC in *O.niloticus*

Malafose con.ppm	No. of alive fish	No. of dead fish	a	b	a*b
0,0	10	-	0	0	0
0.5	9	1	0.5	0.5	0.25
0.75	7	3	0,5	2	1
1	4	6	0.75	4.5	3.375
1,25	3	7	1	6.5	6.5
1,50	-	10	1,25	8.5	10.625
					∑ a*b=21.75

Half lethal concentration of malathion = Highest conc. – $\sum a \times b / n$

LC50= 1.5 – 21.75/10 = 1.0125 ppm ld2.025

Where:

a: Constant factor of difference between groups.

b: Mean value of dead fish between each two successive groups.

n: Number of fish in each group

Following the preliminary experiment with different concentrations (0.75, 1.00, 1.25, 1.50 and 1.75 mg/l) of malafos commercial grade with 57% active ingredient. Fish were exposed for a period of 96h to desired concentration and monitored for determination of LC50 , mortality at 24h/48 and 96h,recorded ,dead fish were removed immediately after death and their numbers registered. Blood was collected from the caudal vein . samples were analyze for RBCs, Hb, Hct and WBC .

Acute Toxicity

Malafos is known as potential toxic pollutant contaminating aquatic ecosystems. The effects of Malafos were studied to determine the 96 h LC₅₀ value on Nile tilapia ,on behavioral changes during test and to investigate hematological parameters of fish exposed to sublethal Malafos concentrations. The 96 h lethal concentration and half LC₅₀ determined with definitive test by the static renewal bioassay method of(J. T Litchfield and Wilcoxon(1949). After finding the LC₅₀, fish exposed to 1/4 and ½ of LC₅₀ then the fish were removed to untreated fresh water with oxygen for 4weeks as arecovery period blood was collected from the caudal vein weekly The one way analysis of variance (ANOVA) was applied to test the t

RESULT

Nile tilapia were exposed to 96 h-LC₅₀ concentration of Malafos 57 EC for 96 h. the results revealed that the 96 h-LC₅₀ was 1.0125 ppm (Table 1)

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However, in the present investigation, the 96 h LC50 value is (1.0125 ppm) of Malafos 57 EC for *O. niloticus*. The results revealed a different mortality rate of fishes which ranged from 0 to 100 % death started less than 30 minutes from the beginning of the tests, showing that the mortality rate was totally dose and time dependent. Fish exposed to malafos started exhibiting behavioral changes and clinical symptoms after 1h hours, they showed loss of equilibrium, irregular erratic and darting swimming movements hyper excitability gasping and accumulated beside the aerater, loss of equilibrium paralysis, sinking of the fishes to

the bottom before death. In addition discharge of mucus, haemorrhage from gills and the dead animals showed blood clots on the mouth and gills. Haematological parameters of *O. niloticus* exposed to malafos treatment resulted in significantly lower values ($P < 0.05$) of RBC, Hb and Ht% compared to those of the control group. while there were significant increase in WBC count, table(2) in contrary to the values of haematological parameters in control group. when fish transferred to fresh water for recovery after exposure to malafos show a very significant recovery in RBC count, Hb and Hct% table(3).

Table2. Effect of malafos on haematological parameters

parameter	control	2days	4 days
RBCs($10^6/\text{mm}^3$)	2,56±0.38	1.7±0.22	1.2±0.1
WBC ($10^3/\text{mm}^3$)	0.86±0.02	2.63±0.23	-
Hb gm/100ml	8.1±0.89	5.8±0.53	5,31±0.13
Ht %	24.6±3.1	16.2±1.57	13±0,12

Data as means ± SE - significantly different ($P < 0.05$)

Table3. Effect of sub-lethal concentrations of malathion on haematological parameters for 4 weeks and arecovery period

parameter	Control	1week	2weeks	3weeks	4weeks
RBCs($10^6/\text{mm}^3$)	2,56±0.38	1.3±0.19	1.7±1.0	2.1±1.1	2.16±0.76
Hb gm/100ml	8.1±0.89	6.1±1.2	6.87±0.60	6.98±1.0	7.5±0.79
Ht %	24.6±3.1	20.9±1.35	21.5± 1.45	23,02±1.2	22.5±1.8

Data as means ± SE - significantly different ($P < 0.05$)

DISCUSSION

The present study showed that the 96h-LC50 of Malafos 57 EC for *O. niloticus* was (1.0125ppm). the results show that malafos is highly toxic to Nile tilapia this might be due to the high toxic potential of the Malafos 57 EC and variation in size of the fish used and environmental factors, such as temperature. These values were relatively high when compared with (Subburaj A.2018) 0.5925±0.0625 /l. (G.V. Venkataraman and P.N. Sandhya Ran; 2013) 1ppm for *Clarias batrahus*. The result is relatively less than the (Naosekpan Sharmila and Gupta Abhik (2013). 9 mg l-1, (Khalid Abdullah Al-Ghanim; 2012) found that the LC50 value as 1.06mg/l. (Josephine O. Boateng et al, 2006) recorded LC50 value of 15.47 µg/l obtained indicates that Deltamethrin is highly toxic to *O. niloticus*. (Aysel Caglan Karasu Benli Ayhan Ozkul; 2010) recorded 0.84 (0.68–1.15) mg/L. (Ehsan H. Abu Zeid and A.L. Shimaa A. Khalil; 2014) The estimated 96 h LC50 was 4.7 mg/L (Sadia Sharmin; 2014) The LC50 value of common carp during the 96 h of exposure was 15.24 mg/L In this study fish show behavioural change while the control groups showed normal behavior during the test

period. A similar observation was also recorded in previous studies. (A. Caglan Karasu et al; 2009), (B, Anbujothi, et al, 2017) recorded that the fish (*Labeo rohita*) showed ungraceful behavior. (Christopher Didigwu Nwani ,et al ;2013) record that fish exposed to various concentrations of the pesticide showed uncoordinated behavior such as erratic and jerky swimming, attempt to jump out of water. (Josephine O. Boateng et al ;2006) On initial contact, the test fishes became inactive, laid down on their sides or remained in a vertical position. (Ehsan H. Abu Zeid and A.L. Shimaa A. Khalil; 2014) Fish of FNT acute exposure exhibited shifts in behavior in the current study which differs depending on dose of FNT exposure. Deviations in behavior were more intense in high exposure level compared with low one. The values of Hb, RBCs and Hct were significantly decreased. WBCs count was increased in the present study, this may be it act as a defence lines. these result match with the result recorded by : (Sadia Sharmin et al ; 2014) recorded that The values of Hb, RBCs and Hct were significantly decreased indicated the failing of hematopoietic system. WBCs count was increased in the present study. The increase

in WBCs count can be correlated with an increase in antibody production. (Lindalva Pereira et al; 2013) Hct presented asignificant decrease, while the parameters Hb, HCM and MCHC presented asignificant decrease indicating ananemic condition. (Heba S. Hamed; 2015) RBCs, Hb, Ht values indicated marked decrease, while the WBCs showed significant increment.

CONCLUSION

The present study concludes that malafos 90h-LC50 for *O.niloticus* was (1.0125ppm).and it is high toxic to the fish. From the present study it can be concluded that expousure of *O.niloticus* to malafos resulted in a significant change in behavior and haematological parameters which causes severe physiological problems, leading to the death of fish. this fact should be taken into consideration when pesticides used for pest control in the agricultural fields surrounding the water bodies.

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