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## Acute Toxicity of Malathion on *Daphnia magna* Straus, 1820

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**Abstract:** The purpose of this study was to determine the acute toxicity and effect of malathion on *Daphnia magna* which was used as test animals. Daphnids were exposed for 96 h to malathion concentrations ranging from 0.001-4 ppm and death daphnids were counted 24, 36, 48, 72 and 96 h later under a stereo microscope. The values of lethal concentration (LC<sub>10,50,90</sub>) and Lethal time (LT<sub>10,50,90</sub>) associated 95-99% confidence limits were calculated. Mortality rate between each dosages and mortality rate between 24-96 h depended on time were correlated. LC<sub>50</sub> values were changed between 0.740-0.015. LT<sub>50</sub> values were found between 110 00'-40 24'. Mortality ratios of *Daphnia magna* treated with malathion during 96 h period was found significant at 95-99% level (p< 0.001-0.005).

**Key words:** Malathion, *Daphnia magna*, lethal concentration, lethal time

### INTRODUCTION

Pesticides may reach the fresh waters, rivers, lakes and streams in a variety of ways. They may be applied to the water bodies for the control of undesirable aquatic fauna or aquatic stages of insects of public health importance. They may also make an incidental impact as a result of aerial application of insecticide for control of terrestrial pests in aquaculture and forestry in surrounding areas. They may also reach lakes and rivers by aerial drift or by run of from land<sup>[1]</sup>.

Malathion was recognized as the first organophosphorus insecticide with high selective toxicity. It was considered to be suitable for same uses of DDT and is now one of the most widely used organophosphate insecticides in the world. Malathion is a non-systemic, widespread insecticide that is used mostly as a general purpose insecticide for control sucking and chewing insects on fruits and vegetables and for controlling mosquito and flies. It is also used to eliminate fish ectoparasites. It could contaminate water supplies as aerial spraying to water bodies for controlling mosquitoes and flies, disposing the waste materials and containers near water supplies or emptying the waste material into sewer drains, or directly using malathion to eliminate the parasites of fish and surface drifts from agricultural area. In aquatic systems malathion first effects the first step of food cycle zooplankton and then fish and finally accumulate in the tissues of animals<sup>[1,2]</sup>.

In aquatic systems malathion can cause many problems on animals such as; altered temperature,

affecting migration and behaviour; inappropriate reproduction timing, decreased growth, skeletal deformities, abortion; decreased enzyme activities; egg production abnormalities; decreased AchE (Acetyl cholinesterase) damaged gills and impaired antibody responses<sup>[2]</sup>.

In measuring the effects of industrial effluents and metallic poison, one of the most widespread microcrustaceans, the water flea, *Daphnia*, has been recognize as particularly suitable indicator species for laboratory tests. As it is one of the most abundant invertebrates in most of the aquatic habitats, always available in numbers and easy to maintain in laboratory culture, it seems the obvious representative of the microfauna in general and one whose role as an indicator of pollution would prove complementary to that fish<sup>[3,4]</sup>.

The purpose of this study was to determine the acute toxicity and effect of malathion on *Daphnia magna* Straus, 1820.

### MATERIALS AND METHODS

**Daphnia culture:** *Daphnia magna* obtained from Fisheries Research Institute (Kepez-Antalya) and have been maintained in culture in our laboratory. The culture acclimated at under 20°C under 14/10 light/dark cycle Daphnids were fed with *Chlamydomonas* sp. The culturing period for one generation was 4 weeks before testing. They showed no sign of stress such as high mortality, presence of male and delay in production of first brood, discoloured animals, etc.

**Test chemicals:** Malathion (CH<sub>19</sub>O<sub>6</sub>PS<sub>2</sub>) was used as test chemical. After many concentrations trial, 13 concentrations was decided for application. The test concentrations of malathion were arranged in a geometric series between 0.001-4 ppm (0.001, 0.002, 0.004, 0.008, 0.016, 0.032, 0.064, 0.125, 0.25, 0.5, 1, 2 and 4 ppm). For each treatment, five replicates were used.

**Toxicity tests:** Daphnids were placed into test tubes (each 10 individual) containing 10 mL water. During the experiment period daphnids were not fed. The tubes acclimated at 20°C under 14/10 light/dark cycle during the study. A control group was used for each treatment.

Each malathion concentration were put into the tubes with Microlit VVCS-10 micropipet and shaken carefully. The number of *Daphnia magna* in each tubes were counted under a stereo microscope after 24, 36, 48, 72 and 96 h later, the mortality rate of each group were determined. Toxicity tests were designed by Ward and Parrish<sup>[5]</sup>.

**Statistic and quantitative structure activity relation-ship analysis:** The values of lethal concentration (LC<sub>10,50,90</sub>) and lethal time (LT<sub>10,50,90</sub>) associated 95-99% confidence limits were calculated by appropriate statistical methods; either binomial, moving average or probit. In the toxicity tests a packet of computer programme (probit) was also used. The values were correlated with logarithm of concentrations. The results were converted to mg L<sup>-1</sup> (ppm). Mortality rate between each dosages and mortality rate between 24-96 h depended on time was correlated<sup>[6-8]</sup>.

## RESULTS AND DISCUSSION

**Effect of malathion concentrations on *Daphnia magna*:** Before exposing daphnids with malathion, the movement and swimming activity of the individuals were seemed to be normal. After treated with malathion, especially in the high concentrated tubes, the swimming activity of Daphnids were changed. They moved faster than the normal conditions, after a time later, the movement of antenna and limbs become slow, deformation of carapace observed, their swimming behaviours changes, in high concentrated tubes daphnids were loosed their original colour.

**The lethal concentrations of malathion:** The lethal concentration of malathion at 24 h could not be defined. Thirty six hour LC<sub>10</sub> value of Malathion was 0.369, where as 96 h LC<sub>50</sub> value was 0.003 ppm. Thirty six hour LC<sub>50</sub>

Table 1: Lethal concentrations (LC<sub>10</sub>, LC<sub>50</sub>, LC<sub>90</sub>) of Malathion depending on time

Concentrations (ppm)	Time (h)				
	24	36	48	72	96
LC <sub>10</sub>	-	0.369	0.022	0.009	0.003
LC <sub>50</sub>	-	0.740	0.120	0.048	0.015
LC <sub>90</sub>	-	1.477	0.619	0.262	0.076

Table 2: Lethal time (LT<sub>10</sub>, LT<sub>50</sub>, LT<sub>90</sub>) of Malathion concentrations

Lethal time (h)	Concentrations (ppm)						
	0.008	0.016	0.032	0.064	0.125	0.25	0.5
LT <sub>10</sub>	77 22'	53 14'	42 42'	40 05'	38 34'	34 05'	25 44'
LT <sub>50</sub>	110 00'	107 20'	70 5,0'	63 30'	58 29'	46 16'	40 24'
LT <sub>90</sub>	166 37'	164 34'	112 15'	97 03'	87 02'	64 31'	61 31'

Table 3: The correlation coefficients, Standard Error (SE), Degree Free (DF) and levels of significant (p) between mortality ratio and concentrations of malathion

Malathion concentrations (ppm)	Correlations of exposure times	SE	DF	p
0.001	-	-	-	-
0.002	-	-	-	-
0.004	0.789	0.355	3	>0.05
0.008	0.828	0.324	3	<0.05
0.016	0.923	0.223	3	<0.05
0.032	0.988	0.089	3	<0.01
0.064	0.985	0.099	3	<0.01
0.125	0.982	0.109	3	<0.01
0.25	0.871	0.284	3	<0.05
0.5	0.852	0.302	3	<0.05
1	0.746	0.385	3	>0.05
2	0.604	0.46	3	>0.05
4	-	-	-	-

Table 4: The correlation coefficients, Standard Error (SE), Degree Free (DF) and levels of significant (p) between death ratio and exposure times of malathion

Exposure times (h)	Correlations of Malathion concentrations	SE	DF	p
24	0.629	0.234	11	<0.05
36	0.789	0.185	11	<0.01
48	0.945	0.099	11	<0.01
72	0.955	0.09	11	<0.01
96	0.923	0.116	11	<0.01

value was 0.740 ppm, 96 h LC<sub>50</sub> value was 0.015; 36 h LC<sub>90</sub> value was 1.477 and 96 h LC<sub>90</sub> value was 0.076 ppm (Table 1).

**Lethal time of malathion:** LT<sub>10</sub> was 77 22' at 0.008 ppm, while this time was determined 25 44' at 0.5 ppm. LT<sub>50</sub> was 110 00' at 0.008 and 40 24' at 0.5 ppm. LT<sub>90</sub> 166 37' at 0.008 and 61 31' at 0.5 ppm (Table 2).

Correlation analysis applied to each concentration. In the concentration of malathion between 0.001-0.002 no death was observed at the end of 96 h. In 4 ppm concentration all daphnids were died immediately after malathion treatment. No significance in the relationship between mortality ratio and time according to 95% confidence limit (p>0.05) was found in 0.04, 1, 2 ppm. In the other concentrations the relationship between

mortality ratio and time was found significant according to 95-99% confidence limit ( $p < 0.05$ - $p < 0.01$ ) (Table 3).

Death ratios of *Daphnia magna* treated with malathion during 96 h period was found significant at 95-99% level ( $p < 0.001$ - $0.005$ ) (Table 4).

Rassoulzadegan<sup>[9]</sup> determined the effect of malathion on *D. magna* as immobility of antenna and limbs, deformation of carapace, changes in swimming behaviour, colourless. The same changes were also observed in this study.

Generally EC<sub>50</sub> values of malathion on *D. magna* was tested<sup>[9-11]</sup>. In this study LC (Lethal concentration) and LT (lethal time) of malathion was determined.

Forty eight hour EC<sub>50</sub> value of malathion was determined as 0.002-0.004 ppm<sup>[9]</sup>. However Cope<sup>[10]</sup>, found that 48 h EC<sub>50</sub> of malathion on *Daphnia* was 2 ppm. Canyurt<sup>[14]</sup> reported that, this value was 0.0009 ppm for 50 h; Lilius *et al.*<sup>[11]</sup> observed LC<sub>50</sub> value as 0.353 ppm. In the present study 48 h LC<sub>50</sub> value was determined as 0.120 ppm. This value was higher than the results of others given above. In the most studies only EC (effective concentration) was observed, but in this study lethal concentration values were determined.

A part from *Daphnia*, the effect of malathion on some other organism was observed in some studies. Twenty four hour EC<sub>50</sub> value was determined as 0.00318 ppm, in *Ceriodaphnia dubia*<sup>[12]</sup>, 0.0027-0.0043 ppm, in *Culex pipiens*<sup>[13]</sup>, 0.5 mg L<sup>-1</sup> in *Mugil capito*<sup>[14]</sup>. In this study lethal concentrations could be calculated after 36 h later.

Post<sup>[15]</sup> reported acute toxicity (96 h LC<sub>50</sub>) of malathion to some fish species such as fathead minnow (9.0 mg L<sup>-1</sup>), largemouth bass (0.285 mg L<sup>-1</sup>), rainbow trout (0.17 mg L<sup>-1</sup>). All these studies shows that *Daphnia* are more sensitive to malathion than fish (96 h LC<sub>50</sub> value 0.015 ppm). Sayım<sup>[16]</sup> found that *Daphnia* are also more sensitive to malathion than *Rana ridibunda*.

In conclusion, Malathion is among widely used pesticides in world. It can contaminate water supplies easily and can affect aquatic organisms. It should not be allowed to contaminate water supplies, because it can affect fish and aquatic invertebrate. Waste materials and containers should not be disposed near water supplies, nor should waste materials be emptied into sewer drains. Instead of chemicals, biological methods could be used for controlling of mosquito and flies in aquatic habitats.

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