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Research Article

Toxicity Studies in Three Indian Major Carps under Stress of Malathion

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Abstract: Malathion is the important pesticide used in Indian conditions. This pesticide in turn through rain wash goes to water ecosystem and affects aquatic fauna especially fishes. Indian carps are major source of food in most of the population. It is necessary to study the toxicity standards and bioassay in Indian major carps under stress of malathion.

Keywords: *Catla catla, Cirrhinus mrigala, Labeo rohita,* Pesticide, Malathion, LC_{50}.

1. Introduction

The most common fishes from Indian point of view are *Catla catla*, *Cirrhinus mrigala* and *Labeo rohita* which are mainly linked to food industry. All these points indicate a sharp study of the vital systems such as toxicity evaluation in these fishes showing the effects of organophosphates of which malathion in the present study to show the activity of these harmful pesticides. Our surroundings have always affected our lives in every way. We need air, water, food etc. for survival and good health but the modern era of industrialization produces various contaminants in our surroundings and add them to air, water and food products. They harm us at every level viz. physiological, anatomical, biochemical, haematological and psychological. The chemistry and mechanism of action of the organophosphate insecticides have been different from those of the organochlorine materials. Organophosphorus compounds merit consideration as they are being used more frequently in place of the organochlorine compounds. The majority of organophosphorus compounds are derivatives of phosphoric acid or the sulphur analogues of phosphoric acid. The phosphoric acid esters used in the synthesis of insecticides of these are prepared industrially by the reaction of phosphoryl chloride with alcohols. These organophosphorus compounds exhibit high insecticidal action against a wide variety of species. This is not really an asset because these compounds are highly toxic to vertebrates as well as to insects and as a consequence, is much more dangerous to the workers using them. The insecticides are mainly used for ornamental plants and shrubs that are not consumed as animal or human food. The ester groups are readily hydrolyzed by the esterase enzyme present in mammals but absent in insects. The toxic effect of the organophosphorus insecticides is due to their interference in the transfer of nerve impulse from one nerve cell to the next. When a nerve impulse reaches the end of a nerve cell it triggers the release of a minute amount of the compound acetylcholine. The acetylcholine activates a receptor on an adjacent nerve cell causing it to carry the impulse to the next nerve cell. The acetylcholine is then hydrolyzed to choline and acetic acid by the enzyme cholinesterase. The organophosphorus pesticide binds chemically the cholinesterase so that it can no longer catalyze the hydrolysis of acetylcholine. The resulting excess of acetylcholine hyperstimulates nerves and produces convulsions, irregular heartbeat and chocking in vertebrates. Main problem associated with organophosphorus pesticide is their environmental conversion to more toxic substances.

Fishes are rich in proteins, lipids, minerals and vitamins and form valuable food for growing population and especially for millions of people suffering from malnutrition and undernourishment in India. Fish haematology has been recommended as a possible means of estimating the health status of fish stacks.

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It is well known that the minute fluctuations in the environmental conditions have early effect on the circulating fluid of the poikilothermic animals. After detailed survey of the literature on the research work, it has been found that very little work has been carried out by some previous research workers on few specific freshwater fishes. Therefore, in present study, an attempt will be made to find out toxicological variations of toxicant in various Indian major carps and it is hoped that the present study will make a considerable enhancement to the existing knowledge on toxicology.

Fig. 1. ***Catla catla.***

Fig. 2. ***Cirrhinus mrigala.***

Fig. 3. ***Labeo rohita.***

2. Methodology

In order to estimate the LC_{50} value, the fishes of different experimental sets have been treated with different concentrations of test compounds as per given in Tables. The mortality number of fishes at different time intervals i.e. 24 hrs, 48 hrs, 72 hrs and 96 hrs and percentage mortality for 96 hrs (Table 1-9, Fig. 4-6) have been calculated which was used as final mortality for calculation as per international standards for fishes. The mortality number showed a corresponding increase with the increasing concentrations of the test compounds.

LC_{50} values have been calculated by the log dose/probit regression line method (Finney, 1971). The test doses have been converted to their logarithms for ease of calculation. Empirical probit values corresponding to the percentage mortality have been obtained from standard table (Finney, 1971) and tabulated in the appropriate columns of the respective tables. The empirical probit values have thereafter been plotted against log dose on the graph paper and a provisional line filling the points is drawn. From this line, expected probit values 'Y' are noted for the values of log dose 'X'. The working probit 'y' has been calculated using the following formula:

$$y = y_0 + kp \quad (1)$$

Where y_0 and k are noted from the table for the expected probit Y and p is the percentage mortality.

The weighing coefficient 'n' for each point is also noted from the table (Finney, 1971). Each weighing coefficient is multiplied by the number of fishes used and the products have been taken as 'w'. After this, for each row, the products of wx, wy, wxy, wx^2, wy^2 have been calculated and summed up as $\sum wx$, $\sum wy$, $\sum wxy$, $\sum wx^2$, $\sum wy^2$ respectively and finally, the mean has been calculated by the following formula:

$$\bar{X} = \Sigma wx / \Sigma w \quad (2)$$

$$\bar{Y} = \Sigma wy / \Sigma w \quad (3)$$

The value of 'b' has been calculated by the following formula:

$$b = (\Sigma wxy - \bar{X}\Sigma wy) / (\Sigma wx^2 - \bar{X}\Sigma wx) \quad (4)$$

Regression equation:

$$Y = \bar{Y} + b\,(x - \bar{X}) \quad (5)$$

Values of 'Y' corresponding to the original values of 'X' have been calculated and the regression line is drawn.

The variance has been calculated by the following formula:

$$\text{Variance (V)} = \frac{1}{b^2}\left(\frac{1}{\Sigma w} + \frac{(X-\bar{X})^2}{\Sigma wx^2 - \frac{(\Sigma wx)^2}{\Sigma w}}\right) \quad (6)$$

The fiducial limits with 95% confidence have been obtained by the following formula:

$$m_1 = m + 1.96\ V \quad (7)$$

$$m_2 = m - 1.96\ V \quad (8)$$

Table 1. Mortality rate of *Catla catla* after treatment with malathion at different time intervals.

S.No.	Concentration (ppm)	No. of fishes	Mortality number after exposure time of			
			24hrs	48hrs	72hrs	96hrs
1	0.04	10	0	0	0	0
2	0.05	10	0	1	2	4
3	0.06	10	1	2	3	6
4	0.07	10	1	2	4	8
5	0.08	10	1	4	8	10

Table 2. Survival number and percentage mortality of *Catla catla* after 96 hours of treatment with malathion.

S.No.	Concentration (ppm)	No. of fishes	Exposure time (hrs)	Mortality number	Percentage mortality	Survival number
1	0.04	10	96	0	0	10
2	0.05	10	96	4	40	6
3	0.06	10	96	6	60	4
4	0.07	10	96	8	80	2
5	0.08	10	96	10	100	0

Table 3. Toxicity evaluation of malathion to *Catla catla* specifying fiducial limits.

Experimental animal	Compound	Regression equation	LC_{50} (ppm)	Variance	Fiducial limits
Catla catla	Malathion	Y = 5.52+8.96 (X-0.78)	0.053	0.0005	m_1 = (+) 0.72098 m_2 = (-) 0.71902

Table 4. Mortality rate of *Cirrhinus mrigala* after treatment with malathion at different time intervals.

S.No.	Concentration (ppm)	No. of fishes	Mortality number after exposure time of			
			24hrs	48hrs	72hrs	96hrs
1	0.05	10	0	0	0	0
2	0.06	10	0	1	1	2
3	0.07	10	1	1	3	5
4	0.08	10	2	4	6	8
5	0.09	10	2	3	7	10

Table 5. Survival number and percentage mortality of *Cirrhinus mrigala* after 96 hours of treatment with malathion.

S.No.	Concentration (ppm)	No. of fishes	Exposure time (hrs)	Mortality number	Percentage mortality	Survival number
1	0.05	10	96	0	0	10
2	0.06	10	96	2	20	8
3	0.07	10	96	5	50	5
4	0.08	10	96	8	80	2
5	0.09	10	96	10	100	0

Table 6. Toxicity evaluation of malathion to *Cirrhinus mrigala* specifying fiducial limits.

Experimental animal	Compound	Regression equation	LC_{50} (ppm)	Variance	Fiducial limits
Cirrhinus mrigala	Malathion	Y = 5.34+13.79 (X-0.86)	0.068	0.0002	m_1 = (+) 0.83039 m_2 = (-) 0.82960

Table 7. Mortality of *Labeo rohita* at different time intervals after treatment with different concentrations of malathion.

S.No.	Concentration (ppm)	No. of fishes	Mortality number after exposure time of			
			24hrs	48hrs	72hrs	96hrs
1	0.06	10	0	0	0	0
2	0.07	10	0	1	1	3
3	0.08	10	1	1	3	6
4	0.09	10	1	2	5	9
5	0.10	10	1	4	8	10

Table 8. Survival number and percentage mortality of *Labeo rohita* after 96 hours of treatment with malathion.

S.No.	Concentration (ppm)	No. of fishes	Exposure time (hrs)	Mortality number	Percentage mortality	Survival number
1	0.06	10	96	0	0	10
2	0.07	10	96	3	30	7
3	0.08	10	96	6	60	4
4	0.09	10	96	9	90	1
5	0.10	10	96	10	100	0

Table 9. Toxicity evaluation of malathion to *Labeo rohita* specifying fiducial limits.

Experimental animal	Compound	Regression equation	LC_{50} (ppm)	Variance	Fiducial limits
Labeo rohita	Malathion	Y = 5.49+15.80 (X-0.90)	0.075	0.0002	m_1 = (+) 0.87039 m_2 = (-) 0.86960

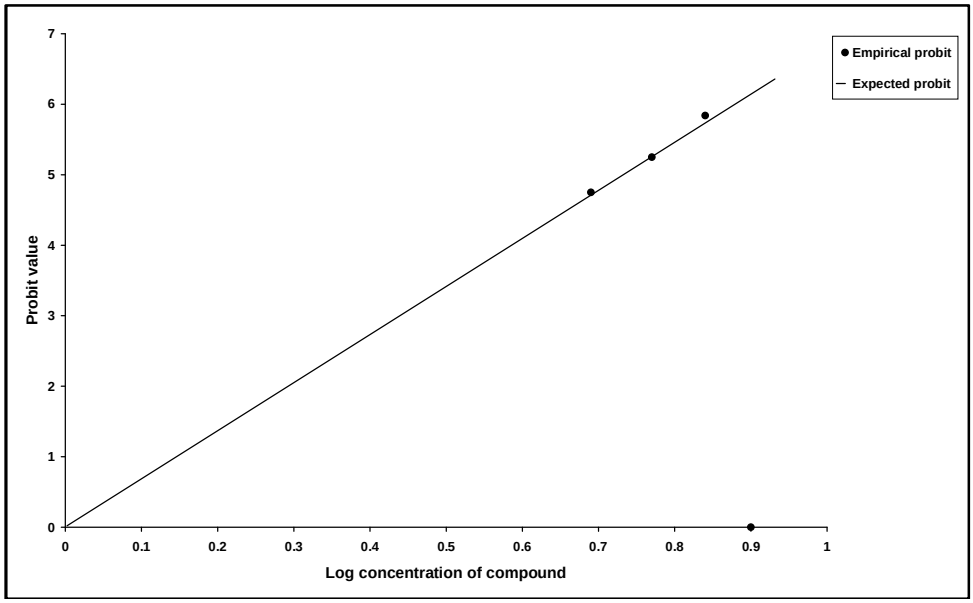

Fig. 4. Regression line for LC_{50} of malathion for *Catla catla.*

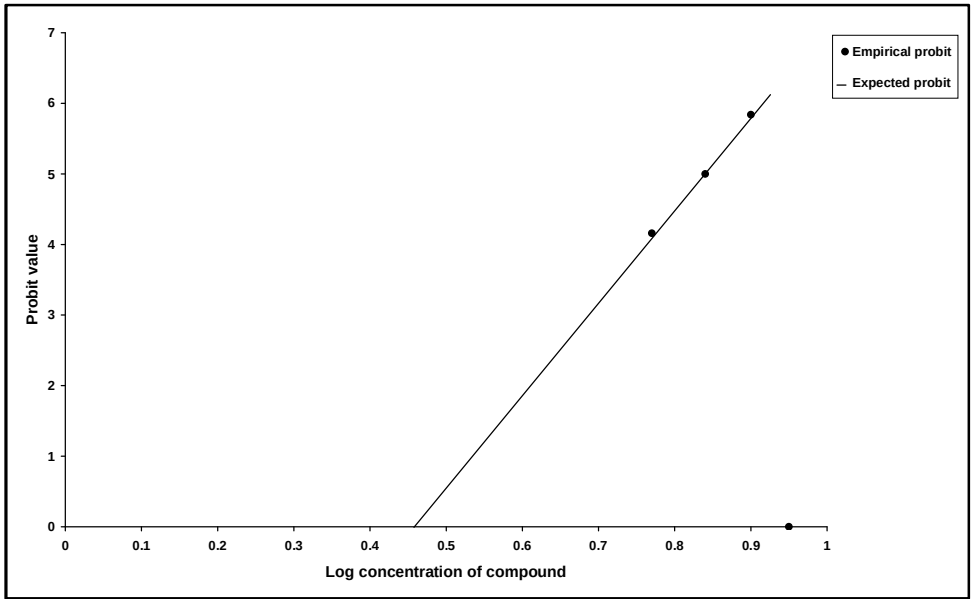

Fig. 5. Regression line for LC_{50} of malathion for *Cirrhinus mrigala.*

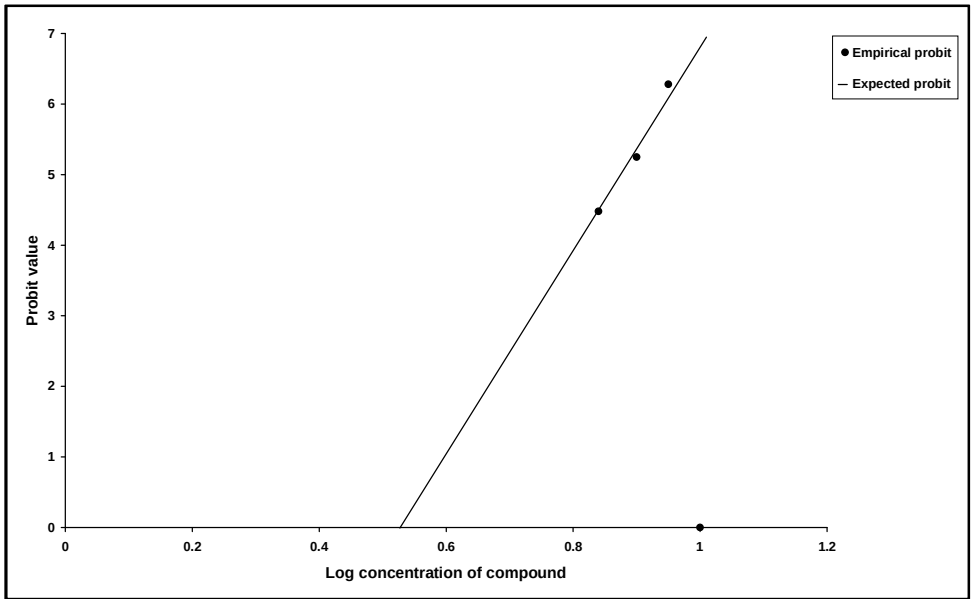

Fig. 6. Regression line for LC_{50} of malathion for *Labeo rohita*.

3. Results and discussion

The LC_{50} value for *Catla catla*, *Cirrhinus mrigala* and *Labeo rohita* were 0.053ppm, 0.068ppm, 0.075ppm for malathion. The sub-lethal concentrations for *Catla catla*, *Cirrhinus mrigala* and *Labeo rohita* were 0.006ppm, 0.007ppm, 0.008ppm for malathion for further studies.

Lethal concentration for 50% mortality is defined as LC_{50} value for a particular species against a particular pesticide. This can be calculated by using different doses against the organism and tested for mortality. Then after a massive statistical calculation, the final LC_{50} value has been estimated which is lethal up to 50% mortality of organism. Then the sub-lethal concentrations are decided by dividing with 10 to minimize the risk of mortality. At last three consecutive concentrations were selected to observe the effect on haematology of the experimental fishes. The LC_{50} value for *Catla catla*, *Cirrhinus mrigala* and *Labeo rohita* were 0.35ppm, 0.46ppm, 0.53ppm for malathion. This reflects that *Catla catla* is most sensitive amongst all them.

The LC_{50} values differ from genus to genus and species to species for the same or different pesticides because of different mode of action and physiology of organism. Environmental factors may also affect the LC_{50} value. Many studies have been done in this regard as Raizada and Rana (1998) reported an LC_{50} value of 0.86mg/L to be highly toxic at 96hrs exposure of *Clarias batrachus* (Linn.) to malachite green. Subramanian *et al*., (2007) studied the toxic effect of heavy metal; chromium on *Clarias batrachus* (Linn.) and reported an LC_{50} value of 2.3401mg/L at 96hrs exposure to be highly toxic. Venkatesan and Subramanian (2007) observed an LC_{50} value of 0.253mg/L at 96hrs exposure of *Oreochromis mossambicus* (Peters) to copper sulphate. The LC_{50} value in the present study is temperature regulated and also depends on water parameters.

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