

## Lethal and Sublethal Toxicity of an Organophosphate Pesticide, Phorate 10G on Fingerlings of *Tilapia Sp.*

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### ABSTRACT

Experiments on bioassay was undertaken to find the toxic effect of 'Phorate 10G' on survival of fingerlings of tilapia. Lethal concentration range of 'Phorate 10G' for tilapia fingerlings was observed to be 0.02 to 0.2 mg<sup>l</sup><sup>-1</sup>. The 96 hrs LC<sub>50</sub> of 'Phorate 10G' for fingerlings of tilapia was found to be at 0.034 mg<sup>l</sup><sup>-1</sup>. Tilapias were exposed to the sublethal concentrations of 0.05 and 0.075 mg<sup>l</sup><sup>-1</sup> for a total period of 30 days. At the end of exposure periods of 10, 20 and 30 days, after every ten days, ten fingerlings were sacrificed for carcass biochemical composition such as moisture content, crude protein, and crude fat and total ash content. The moisture and total ash contents of the tilapia were increased, whereas crude protein and crude fat content decreased as a function of increased the concentration of 'Phorate 10G' and exposure period.

**Key words:** Phorate 10G, Organophosphate, *Tilapia sp.*, LC<sub>50</sub>, Sublethal exposure, Biochemical composition.

### INTRODUCTION

The freshwater aquaculture system constitutes one third of the total fish production of India and the Indian major carps (*Catla catla*, *Labeo rohita* and *Cirrhinius mrigala*) being the dominant species. Mozambique tilapia was imported into India at a time when the quality fish seed, the starting material for aquaculture, was in short supply. Unlike the major carps, tilapia breeds freely and easily produces its seed<sup>7</sup>. Tilapia is a very sturdy fish with more resistant to viral, bacterial and fungal disease than other aquaculture species and tolerate crowding condition with very low dissolved oxygen levels (less than 0.5 mg<sup>l</sup><sup>-1</sup>), which is well below the tolerance levels for most

cultivable fishes, and even high salinity. Tilapia can live and breed even in seawater<sup>14</sup>. The other two species of tilapia, the Nile tilapia and the Blue tilapia are better suited for monosex culture, as they grow faster and mature later than the Mozambique tilapia.

Government of India introduced tilapia in early 1952s for aquaculture purpose and later stocked in impounded waters. Today, tilapia is spread over in all the states of India<sup>8</sup>. Tilapia as an economically important species shares a common food niche and the success of it in competition with the other indigenous spp. is determined by its ability to breed and propagate.

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Tilapia is reported to attain maturity at the age of three months and can breed in every month in the Indian climatic condition have resulting in poor growth rate or even elimination of the indigenous fish species.

The effect of 'Phorate 10G' on *Tilapia sp.* is an important step to understand toxic mechanism of this pesticide. Therefore, the present study is planned to see the effect of 'Phorate 10G' on the survival of tilapia fingerlings.

### MATERIALS AND METHODS

Tilapia fingerlings of appropriate size were collected from the local ponds of Ratnagiri and Dhasai Dam, Indepesca Aquaculture private limited, Thane, Maharashtra state and carefully brought to the laboratory. Fingerlings were treated with 0.05% KMnO<sub>4</sub> solution for two minutes to avoid any cutaneous infection. The disinfected fish stock was kept in 500 liter plastic tank for 10 days to acclimatize under laboratory conditions. The fingerlings were fed twice in a day with proteinous granulated dried commercial fish feed. The faecal matter and other waste materials were siphoned out daily to maintain water parameters in optimum ranges. Everyday, nearly 30 per cent of water from the pool was exchanged. The fish tanks were well aerated, and the physical and chemical parameters were maintained constant in order to acclimatize and provide natural condition to the test fingerlings. The range finding bioassay was conducted following APHA-AWWA-WPCF<sup>2</sup> and Reish and Oshida<sup>15</sup> with fish exposed to a range of sequential concentrations 0.002, 0.02, 0.2, 2.0 and 20 mgL<sup>-1</sup> of 'Phorate 10G'. To determine the lethal concentration (LC<sub>50</sub>) of the 'Phorate 10G', ten fingerlings (average weight 5.518 ± 0.57 gm and average length 6.95 ± 0.44 cm) were released into all glass aquarium tanks (45 x 22.5 x 30 cm) in each tank containing 20 L water and subjected to different concentrations of 0.025, 0.05, 0.075, 0.1 mgL<sup>-1</sup> separately. Thirty fingerlings (4.98 ± 0.632 gm and average length 6.02 ± 0.532 cm) were exposed to sublethal concentrations of 0.01 and 0.015 mgL<sup>-1</sup>. Fingerlings were fed with commercial fish feed, and the tanks were kept well aerated. Ten percent of the medium was replaced after every 24 hrs with fresh

water having same concentration of the insecticide. At the end of exposure periods of 10, 20 and 30 days, 10 fingerlings from each tank were removed and dried for carcass biochemical analysis. The protein content in the sample was estimated by the method of Lowry *et al*<sup>9</sup>. The crude fat was determined by using the method described by Sadasivam and Manickam<sup>16</sup>. The AOAC<sup>1</sup> method was used for determination of moisture and ash content. The experimental data were analyzed by SNK test to determine the significance of the changes from controls<sup>4</sup>.

### RESULTS AND DISCUSSION

The range finding bioassay of 'Phorate 10G' for fingerlings of tilapia, was conducted by taking concentrations of 0.002, 0.02, 0.2, 2.0 and 20 mgL<sup>-1</sup> and 0 as a control. Nil, 20, 60, and 70 percent of mortality were observed when exposed to 0.002, 0.02, 0.2 and 2.0 mgL<sup>-1</sup> respectively. A cent per cent mortality were observed in 20 mgL<sup>-1</sup> concentration. The lethal concentration range of 'Phorate 10G' for tilapia fingerlings was observed to be 0.02 to 0.2 mgL<sup>-1</sup> for a period of 24 hrs (Table 1).

The cumulative average percentage of mortality of tilapia fingerlings exposed for 96 hrs for various concentrations 0.025, 0.05, 0.075 and 0.1 mgL<sup>-1</sup> are given in Fig 1. At the end of 96 hrs exposures, the total average percentage mortality of 40, 60, 100 and 100 was observed in 0.025 mgL<sup>-1</sup>, 0.05 mgL<sup>-1</sup>, 0.075 mgL<sup>-1</sup> and 0.1 mgL<sup>-1</sup> concentrations of respectively.

The 96 hrs LC<sub>50</sub> of 'Phorate 10G' for tilapia fingerlings was found to be 0.034 mgL<sup>-1</sup> respectively. This value differs from LC<sub>50</sub> values calculated by different scientists for different species of fishes and against various compounds. Jaroli and Sharma<sup>6</sup> have reported that the LC<sub>50</sub> of *Channa punctatus* (Bloch) for 96 hrs exposure was 0.365 ppm for dursban. Singh *et al*<sup>17</sup>, have reported that the LC<sub>50</sub> for organophosphate phorate is 0.03 mgL<sup>-1</sup> for *Channa punctatus*. The LC<sub>50</sub> values for 96 hrs of exposure were estimated to be 46.75, 22.95 and 375.26 ppm in *Anabas testudineus*, *Channa punctatus* and *Barbodes gonionotus* respectively by Hossain *et al*<sup>5</sup>, while Auta *et al*<sup>3</sup>, estimated LC<sub>50</sub> values of dimethoate for *Clarias gariepinus* as 39.40 mgL<sup>-1</sup> and for O.

*niloticus* was 20.70 mgL<sup>-1</sup>. Sivaperumal and Sankar<sup>18</sup> reported that the LC<sub>50</sub> for methyl parathion was found to be 10.2 mgL<sup>-1</sup> for *Labeo rohita* of size 75 ± 6g and Ramesh and Sarvanan<sup>13</sup> found it to be 5.28 ppm for *Cyprinus carpio* when exposed to chlorpyrifos.

The sublethal concentration (0.01 and 0.015 mgL<sup>-1</sup>) was decided based on the LC<sub>50</sub> values of 'Phorate 10G' i.e. 0.034 mgL<sup>-1</sup>. After every ten days, ten fingerlings were sacrificed for carcass biochemical analysis (Table 2). Even at lower concentrations of 'Phorate 10G', the values of protein and fat varied significantly (p<0.05) from those of control fish. Significant decrease in the crude protein and crude fat content of fingerlings was observed when exposed to different sublethal concentrations. The present investigation indicated the utilization of all these energy components when fish is under stress. The carcass proximate composition at the end of the 30 days exposure period between control fish and those of exposed to various sublethal concentrations was significantly differed (Table 2). Similar results were reported by Palanichamy *et al*<sup>12</sup>. The moisture and ash levels were significantly increased and the crude protein and crude fat contents were

significantly decreased at various sublethal concentrations of monocrotophos. Protein, lipid and carbohydrate, which constitute the major components of the body, play an important role in body composition and energy metabolism. This is affected by environmental factors like water pollution. Venkataramana *et al*<sup>19</sup>, have recorded a significant decrease in the protein in the gobiid fish (*Glossogobius giuris*), when subjected to higher concentration of malathion (0.5 ppm) for a longer duration (96 hrs). Palaniappan *et al*<sup>11</sup>, have observed a decrease in the nutritive value of muscle of *C. tilapia* fingerlings due to lead toxication. In the present study, crude protein content, crude fat content, moisture content and total ash content showed sensitivity to the sublethal exposure of 'Phorate 10G' over a period of 10 days. The crude protein and crude fat content decreased while moisture and total ash contents increased with increased concentration and exposure period. It thus evident from the present study that increase in concentration and exposure period of "Phorate 10G" have affected the carcass proximate composition depending on the energy requirement of the fingerlings of tilapia.

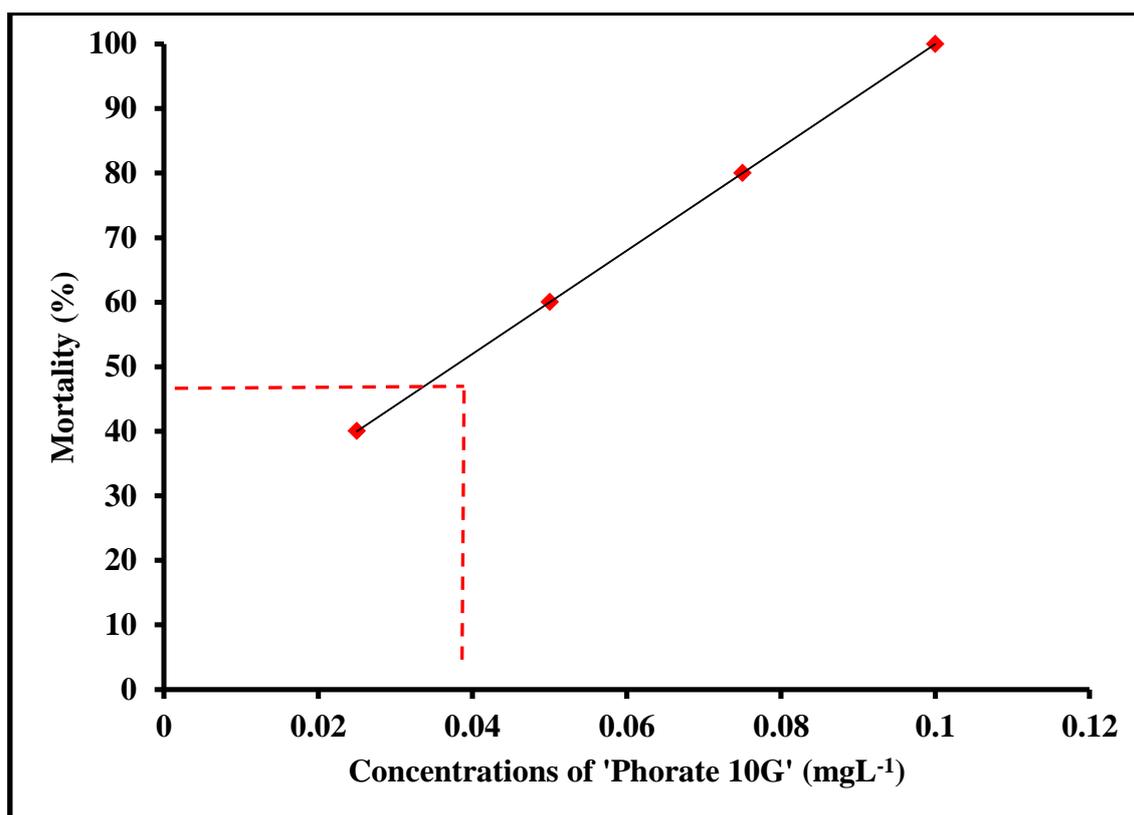
**Table 1: Observation on percentage mortality of tilapia fingerlings (n=10) after every 6 hrs exposed at different concentrations of 'Phorate 10G'**

Test concentrations (mgL <sup>-1</sup> )	Replicates	Mortality of fingerlings observed after each 6 hrs (%)				Cumulative mortality (%)	Average mortality (%)
		6	12	18	24		
Control	1	0	0	0	0	0	0
	2	0	0	0	0	0	
	3	0	0	0	0	0	
0.002	1	0	0	0	0	0	0
	2	0	0	0	0	0	
	3	0	0	0	0	0	
0.02	1	0	20	10	0	30	30.0
	2	20	10	0	10	40	
	3	0	10	0	10	20	
0.2	1	30	20	30	20	100	100
	2	40	20	30	10	100	
	3	30	0	30	40	100	
2.0	1	20	20	40	20	100	100
	2	20	40	10	30	100	
	3	30	20	40	10	100	
20	1	40	20	20	20	100	100
	2	40	30	20	10	100	
	3	40	20	20	20	100	

**Table 2: Biochemical composition of tilapia fingerlings before and after sublethal exposure to 'Phorate 10G'**

Proximate Analysis	initial	Concentration (mgL <sup>-1</sup> )								
		control			0.01			0.015		
		10 days	20 days	30 days	10 days	20 days	30 days	10 days	20 days	30 days
<b>Moisture content</b>	7.9012 ± 0.021 <sup>d</sup>	7.9067 ± 0.0067 <sup>d</sup>	7.9143 ± 0.043 <sup>d</sup>	7.9840 ± 0.002 <sup>cd</sup>	8.4700 ± 0.122 <sup>bc</sup>	8.7300 ± 0.01 <sup>c</sup>	8.8870 ± 0.065 <sup>b</sup>	9.2380 ± 0.003 <sup>a</sup>	9.2617 ± 0.001 <sup>a</sup>	9.2967 ± 0.00 <sup>a</sup>
<b>Crude protein content</b>	73.321 ± 0.001 <sup>a</sup>	73.6307 ± 0.764 <sup>a</sup>	71.9467 ± 0.28 <sup>ab</sup>	71.2967 ± 0.19 <sup>ab</sup>	69.8137 ± 0.271 <sup>b</sup>	67.6007 ± 0.25 <sup>bc</sup>	66.5433 ± 0.14 <sup>c</sup>	66.8007 ± 0.006 <sup>cd</sup>	66.1667 ± 0.034 <sup>d</sup>	66.0767 ± 0.03 <sup>d</sup>
<b>Crude fat</b>	7.8123 ± 0.012 <sup>a</sup>	7.85700 ± 0.02 <sup>a</sup>	7.79367 ± 0.01 <sup>ab</sup>	7.68333 ± 0.035 <sup>b</sup>	7.57000 ± 0.02 <sup>bc</sup>	7.49033 ± 0.01 <sup>c</sup>	7.43733 ± 0.012 <sup>c</sup>	7.34333 ± 0.026 <sup>cd</sup>	7.22633 ± 0.032 <sup>cd</sup>	7.19400 ± 0.003 <sup>d</sup>
<b>Total Ash content</b>	7.8231 ± 0.031 <sup>d</sup>	7.86800 ± 0.04 <sup>d</sup>	8.03533 ± 0.09 <sup>cd</sup>	8.33067 ± 0.03 <sup>cd</sup>	8.51700 ± 0.01 <sup>c</sup>	8.74167 ± 0.05 <sup>bc</sup>	9.09333 ± 0.064 <sup>b</sup>	9.33567 ± 0.04 <sup>ab</sup>	9.90367 ± 0.05 <sup>a</sup>	10.5870 ± 0.124 <sup>a</sup>

Results are given as mean ± SE. Values shown in rows that have different superscripts (a, b, c, d) differ significantly (p < 0.05)



**Fig. 1: 'Phorate 10G' 96hrs LC<sub>50</sub> test for Tilapia**

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