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Seasonal study of acute toxicity of tributyltin oxide (TBTO) on the fresh water bivalve, *Lamellidens marginalis* from Godavari river at Maharashtra, India.

KEYWORDS: *Lamellidens marginalis* , Bis (tributyltin) oxide, TBTO, Seasonal, LC_{50}

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ABSTRACT *The aim of the present study is to investigate LC_{50} values in three seasons of TBTO on L. marginalis. The active substance TBT is highly toxic damaging to non target species. The bivalves have been bioindicators to determine aquatic pollution status. The adverse effect of Tributyltin oxide has been studied on freshwater bivalve mollusk Lamellidens marginalis in summer, monsoon and winter season during June 2014-may 2015. To evaluate the median lethal concentration of TBTO, bioassay was performed in all seasons on fresh water bivalve for 24hr, 48hr, 72hr and 96 hrs. The LC_{50} values in ppm were 1.61, 1.11, 0.85, 0.39 in summer, 2.26, 1.67, 1.27, 0.88 in monsoon and 2.63, 1.92, 1.45, 1.09 in winter season. All values are after 24, 48, 72and 96 hours respectively. Results showed decrease in lethal concentration as exposure period increases and this was observed for all three seasons.*

Introduction

Organotins are widely utilized organometallic compounds, most of which are manmade. For many years tributyltin (TBT) compounds were the favored biocide, it has been used worldwide in antifouling paints for ships and fishing nets, cordage. Therefore, there is increasing interest in the impact of these compounds on the aquatic ecosystems. Organotin compounds are man-made chemical compounds, based on a hydrocarbon structure combined with Sn atom or tin. The most well known organotin is tributyltin (TBT), used widely in aquatic antifoulant paints to prevent the growth of organisms such as crustaceans and molluscans on the hull of ships. Once the organometals are deposited in the environment create a serious threat to global ecosystems. Organometallic compounds are increased risk due to their potential reactivity, bio concentration, bioaccumulation and biomagnification.TBT contamination is still necessary because of the long persistence of TBT in sediment (Chau et al., 1997a; Maguire, 2000; Stewart & Thompson, 1997).

Determination of toxicity of substance is necessary to commercially aquatic forms and water quality management. For the toxicity studies most commonly used methods in aquatic environmental studies with suitable organisms is the bioassay constitute.

According to Elder & Collins, (1991), Freshwater mussels and snails have the potential to be very useful for biomonitoring studies. The reaction and survival of aquatic organism, under toxic conditions depend upon several factors, such as kind, toxicity and concentration of the toxicant and the temperature, salinity, dissolved oxygen, pH and physiological factors such as reproductive cycle and seasons, in addition to the type and time of exposure to the toxicant. (Holden, 1973; McKim *et al.*,1973). Freshwater bivalve mollusks are shown to be year around breeders (Gosh and Ghose, 1972) or the breeding is restricted to year (Mudkhede, 1974) and it is shown to be influence by the changes in environmental factors. The tolerance of *L. marginalis* to different toxicant might be different and it will depend on physicochemical parameters, these parameters always change due to seasonal variation. Abhilash, R. and Prakasam,V. R. (2005)

Bivalves are the bioindicators of metal contamination in water bodies, different species of bivalve have been used to conduct the studies of assessing the toxicity of metals in aquatic system. The present investigation has been planned to assess the effect of tributyltin oxide (TBTO) on mortality of the freshwater bivalve *Lamellidens marginalis*.

Tributyltin oxide is known to be harmful to many, "non-target" aquatic organisms, particularly molluscs (Horiguchi *et al.*, 1997). The pollution of rivers and streams with chemical contaminants has become one of the most critical problems of century (Rane Minakshi and A.Y. Mahajan, 2013). Nikam and Shejule 2015, Rabbito *et al*. 2005, Shejule *et al*. 2006 and Kharat P. S. 2007 had investigated the effects of organometalic compounds on aquatic animals.

The toxicity study is essential to find out toxicants limit and safe concentration, so that there will be minimum harm to aquatic ecosystem. Perusal of literature reveals paucity of information on acute toxicity of tributyltin oxide on freshwater bivalve, *L. marginalis* in all seasons. Hence the present study has been focused to evaluate the seasonal acute toxic effects of tributyltin oxide to freshwater bivalves, *L. marginalis* as bioindicator, of local commercial and environmental importance from Maharashtra state in Godavari river at Paithan.

Materials and Methods

The freshwater bivalves, *Lamellidens marginalis* were collected from the Godavari river at Paithan, 45 km away from Aurangabad city (MH) in different seasons in the year 2014-15. The bivalves were brought to the laboratory and cleaned to remove the fouling algal biomass and mud. The bivalves kept in plastic troughs containing dechlorinated tap water for 4 days to acclimatize to the laboratory

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conditions. Pilot experiments were conducted to find out the range of the toxicity of the toxicant used tributyltin oxide. The chosen range of concentration was such that it resulted in 0 to 100% mortality. 1-ppm stock solution was prepared in acetone, Laughlin *et al*., (1983). The Series of statistic bioassay were conducted under laboratory condition as described by Finney (1971). Acute toxicity tests were conducted over 96 hrs. The experimental troughs containing 5 liters dechlorinated water were used to keep the animals. For each experiment ten bivalves, *L. marginalis* of approximately similar size (60-65mm in shell length) were exposed to different concentrations of tributyltin oxide. After every 12 hours the polluted water was changed by the fresh solution of the same concentration. The behavior and mortality of the bivalves were recorded before each change of water. The resulting mortality was noted in the range of 10 to 90% for each concentration for the duration of 24, 48, 72 and 96 hrs. Each experiment was repeated thrice to obtain constant results. The data collected was analyzed statically by means of probit method on transforming toxicity curve (% mortality vs. concentration), which allows the average median lethal concentration of LC_{50} to be calculated for 24, 48, 72 and 96 hrs. Dead bivalves were counted individually.

Results and discussion

The LC_{50} values were calculated for 24, 48, 72 and 96 hours by Finney's method (1971). The LC_{50} values decreased with increase in exposure period, it was in all seasons. Lethal concentrations of TBTO to *L.marginalis* were different as per seasonal variation. The LC_{50} values, regression results, Chi square, variance and fiducial limits, lethal concentration and safe concentration are shown in Tables 1,2 and 3. The LC_{50} values obtained for tributyltin oxide exposed for 24, 48, 72 and 96 hours exposure were 1.61, 1.11, 0.85, 0.39 in summer, 2.26, 1.67, on ppm 1.27, 0.88 in monsoon and 2.63, 1.92, 1.45, 1.09 in winter season respectively. The result shows that LC_{50} values decreases with increasing periods of exposure of tributyltin oxide and LC_{50} values up to 96 hours exposure period are obtained lowest during summer season. During three seasons toxicity sensation of *L.marginalis* changed due to seasonally variable environmental factors like temperature, light, dissolved contents, P^H, salinity etc. It indicated *L.marginalis* was high sensitive in summer as compare to other seasons and LC_{50} values up to 96 hours exposure are obtained highest during winter showed low toxicity it was less sensitive than other seasons.

Table 1. Relative Toxicity of TBTO to the freshwater bivalve, *Lamellidens marginalis*
Summer season

Time of exposure (Hrs.)	Regression equation $Y=\bar{y}+(X-\bar{x})$	LC_{50} Values in ppm	Variance V	Chi-square	Fiducial Limits		Lethal dose	Safe conc. (ppm)
					M_1	M_2		
24	Y=5.4893X+3.8526	1.618	1.47 X 10^{-3}	0.4542	0.155	0.305	38.78	0.1047
48	Y=3.501X+4.8396	1.111	-3.13 X 10^{-2}	0.3595	-0.02	0.426	53.328	
72	Y=4.2473X+5.2837	0.857	1.53 X 10-^{3}	0.3400	-0.12	0.030	61.732	
96	Y=1.779X+5.70818	0.398	9.49 X 10^{-3}	1.2351	-0.491	0.110	38.208	

Table 2. Relative Toxicity of TBTO to the freshwater bivalve, *Lamellidens marginalis* **Monsoon season**

Time of exposure (Hrs.)	Regression equation $Y=\bar{y}+(X-\bar{x})$	LC_{50} Values in ppm	Variance V	Chi-square	Fiducial Limits		Lethal dose	Safe conc (ppm)
					M_1	M_2		
24	Y=11.6298X+0.8279	2.261	2.7 X 10^{-4}	0.1346	0.329	0.393	54.26	0.18518
48	Y=8.2814X+3.0980	1.679	5.3 X 10^{-4}	1.5092	0.185	0.275	80.59	
72	Y=6.4205X+4.3309	1.272	9.4 X 10-^{4}	0.1980	0.053	0.174	91.584	
96	Y=3.93724X+5.2071	0.885	1.78 X 10^{-3}	0.4347	-0.128	0.036	85.046	

Table 3. Relative Toxicity of TBTO to the freshwater bivalve, *Lamellidens marginalis* **Winter season**

Time of exposure (Hrs.)	Regression equation $Y=\bar{y}+(X-\bar{x})$	LC_{50} Values in ppm	Variance V	Chi-square	Fiducial Limits		Lethal dose	Safe conc. (ppm)
					M_1	M_2		
24	Y=8.0175X+1.56013	2.634	5.1 X 10^{-4}	0.0108	0.387	0.475	63.216	0.2044
48	Y=6.6074X+3.12543	1.921	2.67 X 10^{-3}	0.2453	0.199	0.402	92.208	
72	Y=4.3730X+4.26363	1.458	1.722 X 10-^{2}	0.0231	-0.081	0.433	104.97	
96	Y=6.4392X+4.74701	1.094	9.7 X 10^{-4}	0.0214	0.019	0.102	105.02	

From the result it shows that freshwater bivalve *L. marginalis* is very sensitive to the organometalic compound TBTO and it is highly toxic to *L. marginalis*. Safe concentration of TBTO to the freshwater bivalve *L. marginalis* is 0.10476 ppm in summer, 0.18518 ppm in monsoon and 0.2044 ppm in winter season

Discussion

Survival rate of *L. marginalis* decreased with increasing the exposure period and concentration of TBTO. LC_{50} values had expressed the acute toxicity level for *L. marginalis* exposed to TBTO concentrations. The LC_{50} values obtained for tributyltin oxide exposed for 24, 48, 72 and 96 hours exposure were1.61, 1.11, 0.85, 0.39in summer, 2.26, 1.67, 1.27, 0.88 in monsoon and 2.63, 1.92, 1.45, 1.09 in winter season respectively.

Shejule *et al*., (2006) reported LC_{50} values of the organotin tributyltin chloride exposed to freshwater prawn, *Macrobrachium kistnensis*; up to 96 hours. They showed the LC_{50} values decreased with increase in exposure period. Kharat, (2007) shows the same results of LC_{50} values of the organotin tributyltin chloride exposed to freshwater prawn, *Macrobrachium kistnensis.* Jagtap *et al*. (2010) reported the LC_{50} values of tributyltin chloride exposed to *lamellidens marginalis* for 24, 48, 72 and 96 hours ex-

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posure were 4.6557 ppm, 3.3954 ppm, 2.6535 ppm and 1.7211ppm respectively. They showed that LC_{50} values decreases with increasing periods of exposure of tributyltin chloride. Effects on common oyster larvae exposed to 0.02-100 g/L tributyltin acetate were studied by His and Robert (1985), as a result, in the group of larvae exposed to tributyltin acetate at 0.05 g/L (0.05 g/L in terms of tributyltin chloride) or over, growth was inhibited and deaths were observed within 10 days. Beaumont and Budd (1984) exposed veliger larvae of the mussel (*Mytilus edulis*) to TBTO for 15 days. No larvae survived longer than 5 days in 10 µg/L TBTO, or longer than 10 days in 1 µg/L TBTO. About half the larvae exposed to 0.1 µg/L TBTO were dead on Day 15 (i.e., 15-d LC_{50} approximately 0.1 µg/L TBTO), and most surviving larvae were moribund and had grown significantly more slowly than controls. Nikam and Shejule (2015) reported LC_{50} values of TBTO to freshwater fish *N.botia*. The LC_{50} values were 0.01852, 0.0153, 0.01311 and 0.01099ppm for 24, 48, 72, 96hrs respectively. Gabbott, and Bayne, (1973), have shown that seasonal variations in biochemical composition of molluscs depend on environmental parameters such as temperature and available phytoplankton and factors such as timing of the reproductive cycle and the rate of turnover of stored energy. The effect of tributyltin oxide (TBTO) on freshwater bivalves is minuet different compare to marine organisms and LC_{50} values, safe concentrations are quite different to different species in seasonal variation, physicochemical parameters and available phytoplanktons also effects on impact of TBTO ultimately on LC_{50} values so in the present work it is attempted to seasonal study the effect of tributyltin oxide on survival of freshwater bivalve, *L. marginalis*.

Conclusion

The effect of TBTO on freshwater organisms is quite insufficient compare to marine organisms, so in the present work it is attempted to study the effect of TBTO on survival of bivalve, *L. marginalis*. The present study confirms the toxic potential of this organotins and therefore the use of TBTO in various industries should be controlled.

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