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Toxicity of Tributyltin (TBT) on Zebrafish Embryos

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Abstract

Tributyltin is known as an endocrine disrupting chemicals (EDCs) which are abundant in the aquatic environment. The toxicological research using fish embryos as a model has received considerable interest. In the present study, Zebrafish Fish Embryo Toxicity test (FET) was used to observe the chronic toxicity effects of TBT. FET analysis was carried out for 6 different TBT concentrations (half diluted concentrations) of 100,50,25,12.5,6.2 and 3.1 ng/L. Embryo tests were initiated after 3 hours (~128 cells). Twenty (20 eggs) fertilized eggs were selected for each test concentration and transferred to 24-well plates filled with 2 ml freshly prepared different TBT concentrations (test solutions) and negative control (water). Fertilized eggs were placed in the 24-well plates by using sterilized pipette under the dissecting microscope. The 24-well plates were then covered with self-adhesive foil and incubated at 26° C±1° C for 3 days. Coagulation, heartbeat of the embryo and mortality lethal endpoints were recorded using a dissecting microscope after 8,24,48 and 96 hours. LC50 values were determined by graphically of mortality % vs log TBT concentrations. Triplicates experiments were performed for the FET test. The results revealed that 100% coagulations of the embryos at TBT doses of 50 and 100 ng/L. Therefore, the coagulation is significantly increasing in a dose-dependent manner and TBT might induce coagulation and infertilization of Zebrafish embryo. Heart beat changes were found in a dose-dependent manner with a statistically significant decrease ($p<0.05$) occurred at different TBT doses. LC50 values of TBT for Zebrafish embryo were 19.9,11.7,7.3 and 5.2 ng/L at 8,24,48 and 96 hours respectively. The percentage of mortality was higher in embryos for the trace level of TBT, indicating that embryos are more sensitive to TBT toxicity. Exposure to TBT during 8 hours or more always caused mortality, increasing with longer exposure times. A continuous exposure until 96 hours showed the highest toxicity (LC50=5.2 ng/L). The exposed during the first 8 hours the compound showed less toxicity (LC50=19.9 ng/L) compared to those exposed at 96 hour post fertilization ($p<0.0001$) when the embryos are already hatched. This result suggests that the continuous exposure of TBT cause to the most toxic to Zebrafish embryo with lethal endpoint.

Keywords: Tributyltin, Fish Embryo Toxicity, Lethal concentration, Zebrafish