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Effects of Benzophenone-3 (BP-3); an Ingredient of Sunscreen Cream, on a Common Freshwater Fish Model (*Poecilia reticulata*)

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Abstract

Benzophenone-3 (BP-3) is widely used as a UV filter in personal care products (PCPs) to prevent sunburn, skin damage and as a stabilizer. Due to concerns over endocrine disruption, international regulatory agencies are currently re-evaluating their safety. In this study, the toxic effects of dissolved BP-3 in aquatic environments using a fish model, *Poecilia reticulata* (Guppy) (0.28±0.09 g/ 2.20±0.19 cm), were studied under laboratory conditions. The tested sunscreen is composed of a water-soluble mixture of ingredients containing BP-3 with a labeled percentage of 6% (w/w %). Around 360 adult male *P. reticulata* (weight=0.282±0.001 g; length=2.20±0.01 cm) were collected from a natural habitat that was partially influenced by domestic wastewater effluents (pH=7.6,25.2±0.1 °C) and acclimatized in glass aquaria (45×25×15 cm^3) filled with de-chlorinated water (pH=7.1,25±0.1) for one week (% mortality 1.398). A concentration series of BP-3 (molecular weight: 228.26 g/mol) (0.000[control], 0.1, 0.5,1.0,2.0,3.0,4.0,5.0,6.0,7.0,8.0 and, 9.0 mg/L) was prepared in triplicate using ethanol (added 2mL, for each tank including 0.28213±0.0862 g control) as a solvent in separate tanks containing 6 L of static water with 10 fish. Mortality, abnormal behavior (back swimming), water pH (7.0±0.1), and temperature (25.8±0.1 °C) were monitored for 24-96 hours. Along with the 96 hour toxicity tests, BP-3 levels in dead fish and in the commercial sunscreen were measured using HPLC analysis. According to HPLC analysis, the sunscreen contained 7.79 % (w/w %) of BP-3, exceeding the labelled percentage. The BP-3 concentrations in the fish body in each exposed group were not significant (p>0.05), with each group even with the elevated exposure concentrations. Notably, fish from the control setup showed pre-existing BP-3 (28.391±0.001 mg/kg dry weight), indicating the exposure of collected fish in their natural habitat. Probit analysis determined LC_{50} values for 24,48,72 and, 96 hours as 5.51,3.70,1.68, and 1.19 mg/L, respectively. Similarly, EC_{50} values were 4.71,3.19,2.32 and, 1.42 mg/L, while no observed effect concentrations (NOEC) were 1.04,0.17,0.07, and 0.02 mg/L. The calculated risk ratios (RR) for 24-96 hours were 1.81,4.94,8.37, and 7.92, all of which exceeded the risk level of 1. Hazard Index (HI) values for 48-96 hours of exposure were 1.60,2.09 and, 2.66. These results indicate that BP-3 imposes significant stress and mortality on *P. reticulata* even at relatively low concentrations. Therefore, BP-3 poses a substantial environmental risk when it is released into aquatic ecosystems and recommends replacing BP-3 in sunscreen cream with sustainable alternatives.

Keywords: *Benzophenone-3, Hazard index, P. reticulata, Risk ratio*