

**SYNERGISTIC EFFECT OF WATER HARDNESS AND FLUORIDE ON ZEBRAFISH  
(*DANIO RERIO*) EMBRYONIC DEVELOPMENT**

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Among water quality parameters, hardness and fluoride play a vital role in hatchability, larval growth and survival of fish eggs. Fluoride is mainly derived from the solvent action of water on rocks. Water hardness measures the calcium (Ca<sup>2+</sup>) and magnesium (Mg<sup>2+</sup>) in water. Excessive water hardness levels and fluoride can be toxic and cause oxidative stress in fish. The present study evaluated the synergistic effect of water hardness and fluoride on zebrafish embryonic development. Zebrafish (*Danio rerio*) is an ideal model for studying toxicological and environmental effects on early embryonic development due to its small size, short life cycle, ex-utero development and transparency of the embryo. Following the Sri Lanka standards for potable water, maximum permissible levels of hardness (600 mg/l) and fluoride (1.5 mg/l) were used for the experiment, and the control was filter-sterilized distilled water. Solutions were added into 30 embryos at 2 h post-fertilization (hpf) and retained up to 96 hpf. The experiment was repeated three times using three different batches simultaneously. Variations in hatching rate, hatching time and mortality rate were observed and recorded for the experimental solutions compared to the control. Embryos exposed to the hardness of 600 mg/l, fluoride of 1.5 mg/l and synergistic solution (hardness + fluoride) have recorded a 0% mortality rate and 100% hatching rate. Embryonic hatching time in the three solutions was 60±1.15, 57±3.05 and 78 ±1.15 hpf, respectively, while in the control solution, it was 54±1.15 hpf. No morphological abnormalities were observed other than the hatching delay compared to the control. Thus, the results of the study suggest that hardness and fluoride have a synergistic effect on the hatching time of zebrafish embryos and no critical effects from single exposure under laboratory conditions.

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