

CYTOTOXICITY OF FLUORIDE AND HARDNESS ON VERO, MONKEY KIDNEY CELL LINE: SULFORHODAMINE-B ASSAY

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Recent studies have suggested that the synergistic effect of fluoride and the hardness of drinking water could cause Chronic Kidney Disease of Unknown aetiology (CKDu) in Sri Lanka. Hence, the present study focused on determining the cytotoxicity of fluoride and hardness of drinking water through sulforhodamine-B (SRB) assay using a Vero, a monkey kidney epithelial cell line (ATCC[®] CCL-81TM). Cells were seeded in 96-well plates as 5×10^3 cells/well to determine cell viability. Cells in different wells were exposed to different concentrations of fluoride (0.5, 2.5, 5.0, 7.5, 10.0, 12.5, 15.0 mg l⁻¹), hardness (60, 100, 200, 400, 600, 800, 1000 mg l⁻¹) and fluoride: hardness ratios (0.5:60, 2.5:100, 5.0:200, 7.5:400, 10.0:600, 12.5:800, 15.0:1000 mg l⁻¹). Non-viable cell percentages and CC₅₀ values were determined. Non-viable cell percentage of the cells exposed to the fluoride concentration series ranged from 32.6 to 55.8%, while the cells exposed to hardness concentrations ranged from 17.6 to 54.6%, indicating an increment with the high concentrations. The non-viable cell percentage was increased from 36.6 to 63.8% in the cells exposed to fluoride and hardness ratios. CC₅₀ values of fluoride, hardness alone and fluoride: hardness ratios were recorded as 10.67, 804.04 and 5.74:318.72 mg l⁻¹, respectively. Significantly lower non-viable cell percentages were recorded in cells exposed to fluoride alone than cells exposed to fluoride: hardness ratio ($p < 0.05$); as well, a significantly lower value was observed in cells exposed to hardness alone than cells exposed to fluoride: hardness ratio ($p < 0.05$). Lower CC₅₀ values were observed for the fluoride: hardness ratio compared to the CC₅₀ values obtained for fluoride and hardness alone. Thus, the synergistic effect of fluoride and the hardness of water causes a more significant reduction in cell viability of Vero cells than the effects of fluoride and hardness alone. Hence, the synergistic effect of fluoride and the hardness of drinking water causes cell damage, and it may contribute to the development of CKDu.

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