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Microcystin-LR-induced cytotoxicity and apoptosis in human embryonic kidney and human kidney adenocarcinoma cell lines

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Microcystin-LR (MC-LR) is a potent hepatotoxin, and increasing evidence suggests that it might also induce kidney injury. The aim of the present work was to evaluate the cytotoxicity and possible apoptotic effects of MC-LR on a human embryonic kidney cell line (HEK-293) and human kidney adenocarcinoma cell line (ACHN). Cells were exposed for 24 h to pure MC-LR (1.0–200 μM) and the cytotoxic effects were evaluated by 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) and sulphorhodamine B (SRB) cell viability assays. Cell viability in both cell lines was significantly decreased after treatment with MC-LR at 50 μM for 24 h ($P<0.001$). Moreover, MC-LR-treated ACHN and HEK-293 cells exhibited a marked dose-dependent loss of confluence as judged by phase-contrast microscopy. Similarly, fluorescence microscopic observations following acridine orange–ethidium bromide (AO/EB) staining confirmed that both cell types were undergoing apoptosis after treatment with MC-LR for 24 h. Expression of three apoptosis-related genes, *Bax*, *Survivin* and *p53*, was analysed by quantitative reverse transcriptase PCR analysis. Both *Bax* and *p53* functioned as promoters of MC-LR-mediated apoptosis in ACHN and HEK-293 cells. The *Survivin* gene acted as a suppressor of apoptosis at lower MC-LR concentration (1 μM) and the gene was upregulated at higher MC-LR concentration (10 μM) ($P<0.001$). Significant increases of caspase 3 ($P<0.0001$) and caspase 9 ($P<0.0001$) activity were detected in both cell lines after exposure to MC-LR for 24 h, indicating the MC-LR induces cytotoxicity and a marked apoptosis in both ACHN and HEK-293 kidney cell lines.

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