ROLE OF NO/Ca^{2+}/CaM/MAPK SIGNALLING PATHWAY IN THE MITOGENIC EFFECT OF IL-1β IN C6 GLIOMA CELLS

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Inflammatory factors such as chemokines and cytokines play a critical role in the host defense against disease and injury. Glial cells of the central nervous system (CNS) produce chemokines and cytokines when activated and may be the primary source of CNS disease and injury (1,2). Chronic expression of chemokines and cytokines has been reported to occur in the CNS in a number of neurological disorders including multiple sclerosis/experimental autoimmune encephalomyelitis and Alzheimer's disease (3). To assess a potential role for cytokines in the CNS dysfunction associated with these conditions, we have investigated the effect of IL-1β on the regulation of cell growth. Earlier observations in our laboratory establishing a role for nitric oxide (NO)/Ca^{2+} signalling in IL-1β induced pyrogenic effect (4), prompted us to study this signalling in the IL-1β regulation of cell proliferation. Pretreatment of C6 glioma cells with different doses of IL-1β, resulted in a dose-dependent increase in cell growth. Data showed that both unspecific [N-ω-nitro-l-arginine methyl ester (L-NAME)] and selective {N-[[3-(aminomethyl)phenyl]methyl]-ethanimidamide dihydrochloride (1400W)} iNOS inhibitor, significantly reversed the proliferative response induced by IL-1β. On the contrary, the guanyl ciclase inhibitor, 1H(1,2,4) oxadiazole (4,3-a) quinoxalin 1-one (ODQ), failed to reverse this effect. Either preventing release of Ca^{2+} from endoplasmic reticulum with ryanodine plus 2-Aminoethoxydiphenylborane (2APB) or inhibiting calmodulin activity with N-(6-Aminohexyl)-5-chloro-1-naphthalenesulfonamide hydrochloride (W7), antagonised the mitogenic effect of IL-1β. Regulation of ERK (extracellular signal-regulated protein kinase) activity by IL-1β was also investigated. Data showed that IL-1β induced ERK phosphorilation in a dose dependent manner and this effect was antagonized by L-NAME, 1400W, ryanodine plus 2APB and W7. All together these data indicated that IL-1β upregulated cell proliferation by a mechanism which was cGMP-independent and occurred via the NO/Ca^{2+}/calmodulin/ERK signalling pathway.

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