(CHL) cells with IC50 of 12.8 mM. NOCF induced apoptosis of CHL cells, which was demonstrated by morphological changes, DNA fragmentation and flow cytometric analysis. Treatment of CHL cells with NOCF induced significant G2/M cell cycle arrest without any change in the level of p53 protein. Caspase-3, an executioner of apoptosis was also activated by the treatment of CHL cells with NOCF. The concentration of NOCF inducing apoptosis was so low that we speculate about the environmental significance of these pesticides.

[OA-8] [04/20/2001 (Fri) 15:15 - 15:30 / Room 1]

Hypoxia-induced Neuronal Cell Death Is Caused by Increase in the de novo Synthesis of Ceramide Linked to Caspase Activation

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Ceramide is known to be a lipid-derived second messenger in the cell signaling pathway involved in a variety of cellular responses ranging from cell differentiation, cell cycle arrest, cellular senescence, apoptosis to cell survival and cell proliferation in a number of cells. To examine the role of ceramide in hypoxia-induced neuronal cell death, ceramide generation was measured in SH-SY5Y human neuroblastoma cells metabolically labeled with [3H]palmitic acid or [3H] serine. The chemical hypoxia resulted in a rapid increase in ceramide production with subsequent evidence of cell death in SH-SY5Y cells. The inhibitor of ceramide synthase, fumonisin B1, but not L-cycloserine, a serine palmitoyltransferase inhibitor, reduced the hypoxia-induced enhancement of ceramide. Cobalt chloride also upregulated hypoxia-inducible factor 1a (HIF-1a) known to stimulate the transcription of provoked apoptosis followed by elevation of ceramide levels, but did not induce a concurrent decrease in sphingomyelin. C6-ceramide also induced apoptosis in SH-SY5Y cells in a similar kinetic frame. NOE (N-oleoylethanolamine), an inhibitor of ceramidase, and PDMP (DL-threo-1-phenyl-2-decanoylamino-3-morpholino-1-propanol), an inhibitor of glucosyl ceramide synthase, increased the ceramide level and induced DNA fragmentation in SH-SY5Y cells. Cobalt chloride-induced cell death and ceramide production were significantly potentiated by both NOE and PDMP. A bacterial Sphingomyelinase increased ceramide level, but did not induce cell death. This hypoxia-induced neuronal cell death was potently inhibited by an inhibitor of caspases, z-vad-fmk (z-vad-fluoromethylketone). Our results suggest that hypoxia-induced neuronal cell death may be caused by increase in the de novo synthesis of ceramide pathway and subsequent activation of caspase.

Oral Presentations - Field B

[B1. Physiology] [B2. Pathology] [B3. Neuroscience] [B4. Immunology]

[OB-1] [04/20/2001 (Fri) 13:30 - 13:45 / Room 2]

Role of intracellular calcium release in asiatic acid-induced apoptosis in HepG2 human hepatoma cells

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Pentacyclic triterpenoid compounds have various biological effects, such as anti-inflammatory, anti-hyperlipidemia, wound healing and anti-tumor activities. In this study we investigated cytotoxic effects of asiatic acid (AA), a triterpene acid, on human liver cells. AA decreased viability of HepG2 human hepatoma cells in a time- and concentration-dependent manner. Treatment with 40 M AA markedly induced apoptosis in the cells, measured by DNA ladder formation and flow cytometric analysis of hypodiploid DNA stained with propidium iodide and loss of phospholipid asymmetry detected by annexin-V binding. AA also increased intracellular Ca^{2+} level, which was blocked by TMB-8 and dantrolene, intracellular Ca^{2+} release blockers, but not by EGTA, an extracellular Ca^{2+} chelator. Moreover, AA-induced apoptosis was significantly blocked by the co-treatment with TMB-8 and dantrolene, suggesting that intracellular Ca^{2+} release plays an important role in AA-induced apoptosis in HepG2 cells.

[OB-2] [04/20/2001 (Fri) 13:45 - 14:00 / Room 2]

Urushiol Induced Apoptosis and Cell Cycle Arrest in Human Gastric Cancer Cells

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Urushiols are mixtures of olefinic catechols with a n-C15 or n-C17 alkyl side chain which is isolated from the sap of Korean lacquer tree (Rhus vernicifera Stokes). In the previous study, we evaluated the cytotoxic effect of urushiol on MKN-45 and MKN-28 cell lines. Urushiol mediated DNA fragmentation and nuclear condensation only on MKN-45 cells. In the present study we investigated the modulation of the expression of the apoptosis-related proteins after treatment of urushiol on both MKN-45 and MKN-28 cells. In MKN-45 cells the expression levels of Fas and Fas-ligand proteins were increased after treatment of urushiol. The Bcl-2 protein level was not changed but the Bax protein level was slightly upregulated on MKN-45 cells by the treatment of urushiol. In addition, caspase-3 was activated and PARP protein was cleaved on MKN-45 cells by urushiol treatment. In MKN-28 cells Bcl-2 and Bax protein level were not changed by urushiol treatment. Nevertheless, the cyclin-dependent kinase inhibitors, p21WAF1/CIP1 and p27KIP1 proteins, were upregulated on MKN-28 cell by urushiol. And cyclin-dependent kinase 2 and 4 proteins were decreased. The cyclin-dependent kinase inhibitor protein, a key regulatory protein of the cell cycle, may have contributed to cell cycle arrest in the urushiol-treated MKN-28 cells. These data suggested that urushiol induced apoptosis on MKN-45 cells and mediated cell cycle arrest on MKN-28 cells.

Oral Presentations - Field C

[C1. Biochemistry] [C2. Microbiology] [C3. Cell Biology]

[OC-1] [04/20/2001 (Fri) 14:00 - 14:15 / Room 2]

MODULATION OF HUMAN CYTOCHROME P450 3A4 EXPRESSION BY CERAMIDE, A LIPID SECOND MESSENGER