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FIAU is Phosphorylated and Inhibits DNA Synthesis in Isolated Rat Mitochondria. J. M. Colacino, D. M. Horn, L. A. Neeb, J. W. Horn, and F. C. Richardson. Lilly Research Laboratories, Indianapolis, IN. USA

FIAU is a thymidine analog with in vitro and in vivo against ;hepatitis B virus. Toxicity associated with FIAU treatment included clinical signs consistent with mitochondrial dysfunction. To understand further the mechanism of FIAU toxicity, we examined the effect of FIAU on DNA synthesis in mitochondria. Mitochondria isolated from naive rats were dosed in vitro with concentrations of FIAU or FIAU triphosphate (FIAU-TP) ranging from 0.1 μM to 200 μM. A 14% or 32% decrease in mitochondrial DNA synthesis compared to controls was observed when isolated mitochondrial were dosed with 25 μM FIAU or FIAU-TP, respectively. Since phosphorylation of nucleosides is requisite for the inhibition of DNA polymerase, studies were conducted to determine whether isolated rat mitochondria could phosphorylate FIAU. Results using lanthanum chloride precipitation of phosphorylated products and HPLC analysis showed that enzymes present in a mitochondrial lysate were capable of phosphorylating FIAU to form FIAU monophosphate. In other experiments using intact HepG2 cells, FIAU was found to incorporate into mitochondrial DNA and to cause changes in the ultrastructure of mitochondria.

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Polysaccharide Induced-Apoptotic Cell Death of Variuos Human Lymphoid Cells Integrated with Virus Genome

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Marine micro-alga Dinoflagellate *Gymnodinium* sp.A3 caused ubnormal bloomings with pink-red color in the sea called "red tide" which introduced to the death of cultured fish in the sorounded areas. A strain of *Gymnodinium* sp.A3 produced an extracellular polysaccharide containing sulfate and lactate. Bacteria-free strain of *Gymnodinium* sp.A3 yielded 44 mg of the crude polysaccharide per 1L of the seawater enriched medium. The crude polysaccharide was purified by a DEAE-cellulose column chromatography. Component analyses, IR, NMR, and methylation analyses suggested that the polysaccharide had a linear structure composed of C-4 linked β-D-galactopyranosyl residue containing C-2 linked sulfate and C-3 linked L(+)-lactate. The polysaccharide showed strong cytotoxicities for several human leukemic T cell lines integrated with virus genome such as MT-4 and CEM-LOD cells, but it showed lower toxicities for normal leukemic cell lines such as Molt-4 and U937 cells. Although the mechanism of this selective cytotoxicities of this polymer has not yet been elucidated, DNA extracted after the incubation of MT-4 with this polymer showed apoptotic fragmentation, suggesting that it seems probale that this polymer has specific apoptotic effect to the human lymphoid cells integrated with virus genome.

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FLUPIRTINE PARTIALLY PREVENTS NEURONAL INJURY INDUCED BY PRION PROTEIN FRAGMENT AND LEAD ACETATE

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Flupirtine belongs to the class of triaminopyridines and is successfully applied in the clinics as a non-opiate analgesic drug which possesses also muscle relaxant activity. Here we have used primary cortical cells from rat embryos to demonstrate that flupirtne acts also neuroprotective against the toxic effects caused by the prion protein fragment [$PrP^{106-126}$] and lead acetate [Pb]. These two agents display pleiotropic effects on neurons, among them is an activation of the NMDA receptor complex.

At concentrations above 30μM the toxic peptide fragment of $PrP^{106-126}$ causes neurotoxicity as a consequence of apoptotic fragmentation of DNA. Pb is neurotoxic at concentrations above 10μM. Co-administration of flupirtine [10μM] with either one of the two agents resulted in a reduced neurotoxicity. These data indicate that the cytoprotective effect of flupirtine is measurable *in vitro* also against such noxes which show their effects pleiotropically, including the modulation of the NMDA receptor complex.

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Anti-Rotavirus Activity of Pyrimidine Derivatives

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Antiviral Activities of 2,4-diamino-5,6-substituted pyrimidines (NF-series) introduced β-dimethyl-propanol or β-cyclopropyl-propanol into 4-amino group were evaluated against rotavirus (SA-11 strain). NF-1078 (4:amino-dimethyl-propanol, 5:-H, 6:-Cl) and NF-1083 (4:amino-dimethyl-propanol, 5:-NH_2, 6:-Cl) showed ID_{50} of 8.5 - 10.0 μg/ml and 1.0 - 3.4 μg/ml, respectively, though antiviral indeces were almost 1.0. On the other hand, NF-1062 (4:amino-β-cyclopropyl-propanol, 5:-NH_2, 6:-Cl) derived from NF-1083 showed ID_{50} of 1.0 - 2.6 μg/ml, and antiviral index was more than 30. NF-1059 (4:amino-β-cyclopropyl-propanol, 5:-H, 6:-H) and NF1060 (4:amino-β-cyclopropyl-propanol, 5:-H, 6:-H) had no or low antiviral activity, indicating that the amino group at 5-position and chloral at 6-position influence on the antiviral activity.