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Synthesis and Antiviral Activity of 2'-Deoxy-2',2'-Difluoro-D-Ribofuranosyl Pyrimidine Nucleosides J. S. Kroin, J. Tang, J. D. Nelson, D. C. Jones, L. W. Hertel, Lilly Research Laboratories, Eli Lilly and Company, Indianapolis, In. 46285

A series of 5-substituted cytidine and uridine nucleosides containing the 2-deoxy-2,2-difluoro-D-ribose sugar were synthesized and showed exciting *in vitro* antiviral activity. The uridine series showed potent and selective activity against DNA viruses with the 5-methyl, ethyl and bromovinyl uridine analogs being the most active. The cytidine series was even more potent with broad spectrum *in vitro* activity. In particular, the unsubstituted cytidine and 5-fluorocytidine analogs were active against both RNA and DNA viruses without exhibiting toxicity to preformed monolayers. Synthesis of the nucleosides was based on previously reported preparation of 3,5-t-butyldimethylsilyl-2-deoxy-2,2-difluoro-D-ribose,1. Reaction of the bissilyl derivative,1, with methanesulfonyl chloride provided the C-1 mesylate. Glycosylation of the trimethyl silylated uridines and cytidines with the mesylate afforded the blocked nucleosides which were deprotected to give 2'-deoxy-2',2'-difluoro-D-ribofuranosyl nucleosides.

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Determination of Antiviral Activity of Ganciclovir (DHPG) and Antiretroviral Agents Against Human Cytomegalovirus (HCMV) Using a Novel DNA-DNA Hybridization Assay. W. M. Dankner and S. A. Spector, University of California, San Diego

Since many AIDS patients with HCMV disease are treated with DHPG, we have used a rapid DNA-DNA hybridization assay (Diagnostic Hybrids, Inc.) which eliminates cumbersome DNA extraction,blotting, and autoradiography to evaluate the anti-HCMV effect of DHPG with either zidovudine (AZT), ribavirin (Rbn) or 2,3 dideoxycytidine (ddC). In this assay, infected cells are lysed, wicked onto nitrocellulose membranes, hybridized to an ^{125}I-labeled Mbo 1 fragment of HCMV strain AD169, and counted in a gamma counter. ID_{50}s using this technique correlate closely with values obtained with plaque reduction assays (r=0.83). 3 of 5 HCMV isolates were inhibited by 12 uM AZT ($\bar{X}$ = 31%). With 12 uM AZT/DHPG, the DHPG ID_{50} was decreased for all 5 isolates ($\bar{X}$ ID_{50} 4.2 ± 1.3 vs. 3.0 ± 1.2 uM). At 3 uM AZT, there was no effect alone or combined with DHPG. For DHPG/Rbn, the $\bar{X}$ DHPG ID_{50} for 5 HCMV isolates was 3.7 uM ± 0.8 uM vs. 3.4 ± 1.1, 3.6 ± 1.8 and 3.8 ± 1.5 uM when combined with 1, 5 and 10 uM Rbn, respectively. For 4 HCMV isolates, the $\bar{X}$ ID_{50} for DHPG alone and combined with 1 uM ddC was 2.9 ± 0.8 uM vs. 2.7 ± 0.6 uM. Thus using a rapid nucleic acid hybridization assay, we have found that when DHPG is combined with AZT, Rbn or ddC at concentrations achieved clinically that the anti-HCMV effect of DHPG will be either unchanged or enhanced with AZT.