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Inhibitory Effect of 2-Phosphonomethoxyalkyl Derivatives of N6-Substituted 6-Aminopurines on Epstein Barr Virus (EBV)

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We have evaluated the anti-EBV activity of a new series of acyclic nucleoside phosphonate analogues with, as the acyclic side chain either phosphonomethoxypropyl (PMP) or phosphonomethoxyethyl (PME) (attached to the N7 or N9 position of the purine ring) and various substituents in the purine ring. Several compounds of these series were shown to possess antiherpes (HSV-1, HSV-2, CMV, VZV) activity (Holy et al., Antiviral Res. 26, no. 3, p. A231). The anti- EBV activity was assessed by means of an EBV DNA hybridization assay using a digoxigenine-labelled probe (specific to the Bam HI-W fragment of the EBV genome) and by measuring viral capsid antigen (VCA) expression by immunofluorescence after a 7-day incubation period of P3HR-1 producer cells with the test compounds. Acyclovir, Ganciclovir, HPMPC, and azidothymidine, which were included as reference compounds gave selectivity indexes (SI) of >250, 132, 25, and 5, respectively. Several of the new nucleoside phosphonates proved to be potent and selective inhibitors of the EBV replicative cycle and EBV VCA production. The acyclic nucleoside phosphonate analogues 9-(2-PME)-6-dimethylamino-purine, 9-(2-PME)-2-amino-6-benzhydrylaminopurine, 7-(2-PME)-6-dimethylaminopurine, 9-(R)-(2-PMP)-6-(2-dimethylaminoethyl) aminopurine yielded a SI of 91, 28, 55, and 37, respectively.

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A dot blot method for detection of HHV-6 antigens and its utilization for evaluating antiviral drugs

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A simple, reproducible and comparable method for detection of human herpesvirus 6 (HHV-6) antigens was developed using a dot blot assay to evaluate the effect of antiviral drugs. Anti HHV-6 activity against three groups of drugs was investigated by using this method. Phosphonoformic acid (PFA), a nonnucleoside derivatives, 9-[(1-3-dihydroxy-2-propoxy)methyl]guanine (DHPG), 9-(2-hydroxyethoxymethyl) guanine (ACV) and 9-[4-Hydroxy-3-(hydroxymethyl) butyl]guanine (PCV), guanosine analogs and (S)-1-[(3-hydroxy-2 phosphonylmethoxy) propyl]cytosine [(S)-HPMPC)], (S)- cyclic-HPMPC (cHPMPC), HPMEMG, acyclic nucleoside phosphonates (ANPs), were examined. In cord blood mononuclear cells (CBMCs) infected with HHV-6B at MOI of 0.004 $CCID_{50}$/cell, the end-point concentration (EPC, which was determined from a dot blot assay) and EC_{90} values for DHPG, (S)-HPMPC and cHPMPC were approximately 1 μg/ml, essentially similar and highly effective among drugs. Following HHV-6A infection at the same MOI, the EPC and EC_{90} values for these three drugs, ACV and HPMEMG were twice than that for HHV-6B. The EPC and EC_{90} values of PFA for HHV-6A were the same as that of HHV-6B. These results suggest that the susceptibility of enzyme for replication can be different between HHV-6A and B.

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Treatment of Progressive Multifocal Leukoencephalopathy (PML) With Cidofovir in an AIDS Patient

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PML is a rare, demyelinating disease, caused by the human polymomavirus JC, occurring in immunocompromised patients, particularly those with AIDS. There is no established treatment for PML and the disease is usually rapidly fatal. We report here the case of a 30-year old homosexual man who was hospitalized with an history of right hemiparesis progressing since 3 weeks. The patient was also slightly dysarthric. Nuclear magnetic resonance (NMR) of the brain revealed an hypodense lesion within the white matter of the left parietal lobe, highly suggestive of PML. A stereotaxic brain biopsy was performed that confirmed the diagnosis of PML, showing severe demyelinisation, lipid-laden macrophages and reactive astrocytes. The patient was treated by intravenous injection of cidofovir (HPMPC) at 5 mg/kg (total dose 300 mg) every 10 days. On the day of each injection, the patient was prehydrated with 1 liter NaCl 0.9% in one hour and treated with probenecid (4 g). In total, 4 courses of cidofovir were given. The treatment was very well tolerated and biochemistry as well as hematology remained unchanged. Neurological examination showed a slow progressive evolution of hemiplegia. Cidofovir was stopped, and the patient died 2 months later. A PET scan performed 8 days after the second cidofovir infusion showed a clear reduction of the hypometabolic left fronto-parietal area, compared to the initial scan before therapy. In the CSF samples taken 1 h after the cidofovir injection, 0.25 µg/ml HPMPC could be detected by HPLC. About 70% of the HPMPC was recovered in the urine within 12 h after administration of cidofovir. These preliminary results suggest that cidofovir should be further explored in the treatment of PML in AIDS patients, using more frequent injections (i.e. every week) and a systematic follow-up by PET scanning.

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Anti-Influenza Virus Activity of Pyrimidine Derivatives

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Antiviral activities of 2,4-diamino-5,6-substituted pyrimidines (NF-series) introduced 8-cyclobutyl-propanol to 4-amino group were evaluated against influenza virus A (A/PR/8/34, IVA) and B (B/Gifu/2/73, IVB). NF-1056 (5:-NH_2, 6:-Cl) and NF-1172 (5:-CH=NOH, 6:-Cl) showed ID_{50} of 2.5 - 11.5 µg/ml and 0.4 -1.0 µg/ml, respectively. No difference of ID_{50} against IVA and IVB was observed. NF-1057 (5:-H, 6:-Cl) and NF-1075 (5:-H, 6:-H) showed decreased activity against both IVA and IVB as compared with NF-1056, indicating amino group at 5-position and chloral at 6-position are effective for the antiviral activity. Derivatives introduced phenylalkyl into 3'-position of cyclobutyl of NF-1056 had a high anti-IVA and IVB activity; 3'-phenyl-, 3'-benzyl-, 3'-phenetyl- and 3'-phenylpropyl-derivatives (NF-1151, NF-1144, NF-1147, NF-1153, respectively) had ID_{50} of 1.0 - 2.0, 0.06 - 0.1, 0.07 - 0.6, and 0.03 - 0.02 µg/ml, respectively, though cellular toxicity increased in parallel.