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Antiviral Evaluation of A-73209, a Potent, Orally Active Agent Against VZV and HSV Infections. J. Clement[1], J. Alder[1], K. Marsh[1], D. Norbeck[1], W. Rosenbrook[1], T. Herrin[1], C. Hartline[2] and E. Kern[2]. Abbott Laboratories[1], Abbott Park, IL and the University of Alabama[2], Birmingham, AL, U.S.A.

A-73209 is a novel nucleoside analog of oxetanocin with potent and selective activity against VZV, HSV-1 and HSV-2 in a plaque reduction assay using human foreskin fibroblasts. Against five TK^+ strains of VZV, A-73209 had mean EC_{50} values of 0.01μg/ml (range 0.003-0.03) compared to 1.22μg/ml (range 0.6-2.7) for acyclovir. Against HSV-1 isolates, A-73209 with a mean EC_{50} of 0.03μg/ml (range 0.01-0.05) was more potent than acyclovir which had a mean EC_{50} of 0.32μg/ml. The mean EC_{50} of A-73209 against a panel of HSV-2 TK^+ strains was 2.2μg/ml. No cytotoxicity was noted at concentrations up to 100μg/ml. The compound was more effective than acyclovir against murine HSV-1 infections when administered IP or PO. In a murine systemic HSV-1 infection (10 LD_{50}), A-73209 given IP or orally had ED_{50} values of 30 and 142mg/kg compared to acyclovir ED_{50} values of 128 and >500mg/kg. In a lethal HSV-1 mouse infection resulting from intranasal inoculation of 10 LD_{50} (2.8×10^4 PFU), A-73209 produced 100% survivors at oral doses of 62.5mg/kg or more while the ED_{50} of acyclovir was 146 mg/kg with 100% survival produced at a dose of 250mg/kg. Against systemic HSV-2 infections in mice, oral A-73209 was as effective as acyclovir while IP A-73209 (ED_{50} 99 mg/kg) was moderately less potent than acyclovir (ED_{50} 56 mg/kg). A-73209 demonstrated good bioavailability in mouse, rat, dog and monkey with %F values of 50, 39, 100 and 37, respectively. Terminal half-lives following IV administration were 0.70, 0.46 and 0.60 hours in rat, dog and monkey. Oral administration of 10 mg/kg to rat, dog and monkey resulted in Cmax values of 4.70, 14.29 and 3.70μg/ml. A-73209 appears to be a selective, potent and orally bioavailable agent for the treatment of varicella-zoster and HSV infections.

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Inhibition of Herpes Simplex Virus Infection by Antisense Oligonucleotides, *R. Hanecak, V. Driver, B. MacDonald, R. Azad, C. Ford, and K. P. Anderson, ISIS Pharmaceuticals, Carlsbad, CA , 92008, USA .*

Phosphorothioate oligonucleotides complementary to mRNA transcripts encoding essential HSV gene products were examined for antiviral effects in cell culture assays. Antiviral activity was initially determined by 96-well immunoassay (ELISA) using primary human dermal fibroblast (NHDF) cells under serum-free conditions. Dose-response comparisons of antisense oligonucleotides resulted in the identification of 5 compounds with potent anti-HSV activity. All compounds inhibited several HSV-1 strains including acyclovir resistant isolates, and HSV-2. 90% inhibition of infectious virus yield was achieved at a concentration of 3μM. Structure-antiviral activity relationships were determined by ELISA. Sequence comparisons of active oligonucleotides resulted in the identification of a sequence motif common to all 5 antisense oligonucleotides which was required for maximal antiviral activity. However, sequences flanking the motif also contributed to antiviral activity. Time-of-addition experiments showed that phosphorothioate oligonucleotides at a concentration of 12μM nonspecifically inhibited HSV infection only when added with the virus (t=0 hrs). However, antisense oligonucleotides designed to target essential HSV mRNAs inhibited HSV-1 replication when added as late as11 hrs. post-infection. Several oligonucleotides showing activity in HSV viral inhibition assays also inhibited human cytomegalovirus (HCMV AD169) , influenza virus, and human imunodeficiency virus (HIV) suggesting an alternative mechanism of action. The same oligonucleotides did not inhibit human rhinovirus, poliovirus or adenovirus. Active oligonucleotides showed only minimal cytotoxicity in MTT or neutral red uptake assays at concentrations up to 100μM. However, alterations in cell morphology were evident.