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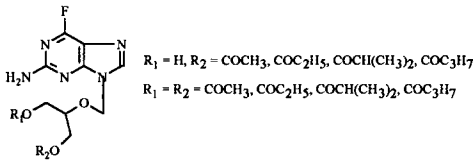
NOVEL 2-AMINO-9-(1,3-DIHYDROXY-2-PROPOXY-METHYL)-6-FLUOROPURINE MONO- AND DIESTERS AS POTENTIAL PRODRUGS OF GANCICLOVIR. D.-K. Kim*, K. Y. Chang, G. J. Im, H.-T. Kim, Y.-B. Cho, and K. H. Kim. Life Science Research Center, Sunkyong Industries, Suwon-Si, Kyungki-Do, Korea

The limited oral absorption of ganciclovir prompted us to search for its oral prodrugs. Previously, we showed that a 6-deoxy-6-fluoro congener of ganciclovir was efficiently converted to ganciclovir by adenosine deaminase (*Bioorg. Med. Chem. Lett.* 1994, 4, 1309). In this study, a series of 2-amino-9-(1,3-dihydroxy-2-propoxymethyl)-6-fluoropurine mono- and diesters have been prepared to increase their oral absorption rates and their efficiencies as prodrugs were evaluated in mice and rats. The ganciclovir concentrations in the blood were measured by HPLC after oral administration of the prodrugs at doses of 100 μmol/kg in mice and 200 μmol/kg in rats, respectively. All prodrugs produced greater amounts of ganciclovir in the blood. The peak plasma concentrations for ganciclovir achieved with monoesters were 3- to 5-fold higher in mice and 16- to 19-fold higher in rats than that estimated for an equivalent dose of ganciclovir. The AUCs for ganciclovir were 3- to 5-fold higher in mice and 12- to 15-fold higher in rats than that of ganciclovir. This study indicates that 2-amino-9-(1,3-dihydroxy-2-propoxymethyl)-6-fluoropurine monoesters are highly efficient ganciclovir prodrugs suitable for oral administration.



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Ganciclovir Resistant CMV: Implications of UL97 and Polymerase Mutations in Cross Resistance to Cidofovir. J.M. Cherrington[1], I.L. Smith[2], R.E. Jiles[2], M.D. Fuller[1], M.S. Chen[1] and S.A. Spector[2]. [1]Gilead Sciences, Foster City, CA, U.S.A.; [2]University of California San Diego, La Jolla, CA, U.S.A.

CMV resistance to ganciclovir (GCV) is a result of a mutation in UL97, polymerase, or both genes. Since cidofovir is an acyclic phosphonate nucleoside analog of 2'-deoxycytidine monophosphate, and does not require UL97 for antiviral activity, cross resistance to cidofovir is observed only as a result of a polymerase mutation. We undertook a study to determine the prevalence of cross resistance to cidofovir among GCV resistant isolates. Of 22 GCV resistant isolates, 11 were found to be cross resistant to cidofovir. Each of these cross resistant isolates was noted to have high level GCV resistance (>6-fold; IC_{50} >30 μM). In contrast, all 11 of the cidofovir sensitive isolates had low level GCV resistance (<6-fold; IC_{50} <30 μM). Genotypic analyses indicated that 10 of the 11 isolates with low level resistance carried UL97 mutations at amino acids 460 or 592–595, as did 9 of 9 isolates with high level GCV resistance sequenced thus far. Additionally, portions of the polymerase genes of 8 of these 11 high level resistant isolates have been sequenced and all 8 show mutations in regions previously implicated in GCV resistance and cross resistance to cidofovir. These data suggest that low level GCV resistance is due primarily to UL97 mutations; these isolates are sensitive to cidofovir. High level GCV resistance is due primarily to a combination of UL97 and pol mutations; these isolates are cross resistant to cidofovir. Continued phenotypic and genotypic analyses on additional resistant isolates are ongoing and will be presented.

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The effects of combining T-cell immunosuppression with antiviral chemotherapy studied in a murine model for cutaneous HSV.

Field, H.J. & Thackray, A.M. Centre for Veterinary Science, Cambridge University, Madingley Road Cambridge, CB3 0ES, UK.

Some of the most successful antiviral chemotherapy has been in immunocompromised patients with severe HSV infections. We have established a murine model in which cyclosporin (Cy) at 50 mg/kg s.c. on alternate days induces immunosuppression. Mice show no toxic effects, but their T-cell function is impaired[1]. Mice were treated with either famciclovir (FCV) or valaciclovir (VACV) orally at 50 mg/kg/day b.i.d. for 5 days commencing on day 1 or 5 p.i. Half the mice were given Cy from days -2 to 16 p.i. Clinical signs, weight-loss, local inflammation, mortality and virus replication in skin and neural tissues were recorded. Fifty and 21 percent untreated mice died by day 16 with and without Cy respectively. No mice treated with FCV died; but in the group treated with VACV from days 5-10, mortality was 25% and 11% with and without Cy respectively. Without chemotherapy, Cy prolonged virus replication in both skin and CNS to the end of observation. Notably, FCV (from day 1-5) cleared virus from skin and CNS although virus was still detectable at the last treatment time. Therapy with VACV, however, did not clear virus permanently and tissues were repeatedly positive for infectious virus during the period 5-13 days p.i. Tissue specimens from mice treated from day 5-10 are currently under investigation. Mechanisms will be proposed for the different effects of the two antiviral compounds on pathogenesis.

[1]**Antimicrob. Agents Chemother. 6, 210-216, 1995.**

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The Design, Chemistry, and Antiviral Activity of a Novel Group of Anti-Herpesvirus Prodrugs.

S. Spruance, M. McKeough, I. Yanachkov, G. Wright, E. Kern, R. Sidwell, D. Smee, M. Yanachkova, A. Glazier. U. of Utah, Salt Lake City, UT; U. of Mass., Worcester, MA; U. of Alabama, Birmingham, AL; Utah State U., Logan, UT; Drug Innovation and Design, Inc. (DIAD), Newton, MA, USA

Current topical therapies for recurrent herpes labialis and genitalis are inadequate. To address this need, we have developed a novel series of acyclovir phosphate and H-phosphonate prodrugs designed to be lipophilic with enhanced potential to penetrate the skin. These prodrugs are based upon a novel class of phospho-esters which undergo C-O bond cleavage via an esterase-triggered elimination reaction which produces the parent compound and by-products of low toxicity. These compounds have shown significant *in vitro* and *in vivo* activity against herpes simplex virus (HSV). One prototype compound, **A7-183,** consists of acyclovir phosphate coupled to 2 equivalents of methyl 3-hydroxy-3-(4-acetoxy-phenyl) propanoate. **A7-183** was evaluated in the dorsal cutaneous guinea pig model of HSV infection. Eight guinea pigs were infected on day 0 with HSV-1 at 4 separate sites per animal. Sites were treated on days 1, 2, and 3 with either 5% acyclovir ointment, ointment control, **A7-183** as a 1.2% aqueous cream, or cream control. Efficacy was evaluated on day 4 by assessment of lesion numbers, area and virus titer at each site.

Measure (mean)	A7-183	Control	Acyclovr	Control
# of sores	32	58	55	56
Total area (mm^2)	106	259	211	253
Virus (log pfu/ml)	3.07	4.49	4.09	4.52

A7-183 exhibited striking antiviral and clinical activity and was superior to acyclovir by all 3 efficacy parameters. (P=.001) **A7-183 cream** is the most effective aqueous-based nucleoside antiviral treatment tested to date in this model. This class of compounds appears to have great promise for the topical therapy of recurrent herpes labialis and genitalis.