13

Phosphonate derivatives of ganciclovir and acyclovir inhibit murine cytomegalovirus (MCMV) infections in cell culture and in mice. D.F. Smee[1], S.T. Sugiyama[1], J.H. Huffman[1], N. Zaveri[2], and E.J. Reist[2]. [1]Utah State University, Logan, Utah, and [2]SRI International, Menlo Park, California USA.

Phosphonate derivatives of ganciclovir and acyclovir were synthesized and evaluated for anti-MCMV activity. Compounds included acyclovir phosphonate (SR3722), and the S-enantiomer (SR3772), R-enantiomer (SR3773) and R,S-enantiomeric mixture (SR3745A) of ganciclovir phosphonate. In C127I cell culture 50% plaque reduction assays, the nucleotides were effective at 4.3 (SR3722), 0.6 (SR3745A), 3.1 (SR3772 and 0.4 (SR3773) μM, whereas ganciclovir was effective at 6.0 μM. Virus yield reduction assays confirmed the results of the plaque assays. The antiviral activity of SR3772 was surprising since it was not phosphorylated by guanylate kinase in test tube reactions, whereas SR3773 was effectively metabolized to its monophosphorylated derivative. The compounds were relatively non-toxic to proliferating C127I cells, with 50% inhibition of cell growth observed at 250-1000 μM. In severe combined immunodeficient (SCID) mice infected with MCMV, doses of 12.5 to 50 mg/kg/day of SR3773 given once daily for a week were far superior to ganciclovir in extending the mean time to death. In contrast, SR3722 at 50 mg/kg/day was only marginally effective relative to the placebo control. A comparison of SR3745A to SR3773 in SCID mice showed that SR3745A was only half as active as SR3773, suggesting that in these animals only one enantiomer (the R-enantiomer) is biologically active. In normal BALB/c mice, SR3772 was effective in completely preventing mortality at 100 mg/kg/day. This compared with equivalent activity of SR3773 at 6.25 mg/kg/day. Overall, these types of guanine analogs appear to be non-toxic and effective inhibitors of MCMV infections. Supported by PHS Grant AI33375-02, NIAID.

14

Treatment of Viral Retinitis with High Dose HPMPC Liposomes

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We have shown that multivesicular liposome-encapsulated HPMPC, in a concentration of 100 g is therapeutically effective against herpes retinitis when injected intravitreally for up to 60 days using the rabbit model. We now show that a concentration of 1000 μg of HPMPC encapsulated in liposomes is non-toxic to the rabbit retina for periods up to 8 weeks. To further evaluate long term efficacy of HPMPC, 17 Dutch pigmented rabbits were injected with 0.1 ml 1000 μg encapsulated HPMPC 8, 12, 16 and 32 weeks prior to transvitreal inoculation of the retinal surface with 10^4 pfu of HSV-1 (ph strain). In each group, 2 eyes of 2 different animals were used as controls. One eye was injected with saline while the other eye injected with virus. ERGs were performed at the baseline and before injection of the virus to assess possible longer term toxicity. Animals were followed up on days 1,3,5,7,14,21 and 28 days by indirect ophthalmoscopy and slit lamp examination. Animals were sacrificed, the eyes were immersed in parafolmaldehyde 4% and studied under light and electron microscopy. Immunostainning for HSV-1 was performed in all eyes injected with virus. ERG tracings were normal in all study groups, but localized pigmentary changes were observed in the area adjacent to the liposomes. No retinitis was observed in any time-point up to 16 weeks, both clinically and histologically. This finding was confirmed by immunostainning. The 32 week time-point is currently being evaluated. These data suggest that high dose liposome encapsulation of HPMPC significantly prolongs the intravitreal antiviral duration of action to over 4 months making this an exciting option for treating HCMV retinitis as HCMV is over 50 times more sensitive to the effect of HPMPC than is HSV

Supported by NIH EYO7366 and the U.C. AIDS task force (WRF).