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Pattern of Resistance Against AZT-Resistant HIV-1 in Human Peripheral Blood Mononuclear cells. R. F. Schinazi,* A. McMillan, D. Cannon, R. Mathes, and C.K. Chu. Department of Pediatrics, Emory University School of Medicine/VA Medical Center, Atlanta, GA, and College of Pharmacy, University of Georgia, Athens, GA, USA.

The development of resistant-HIV variants to various clinically useful antiviral agents could have an important impact on the treatment of individuals infected with HIV-1. Studies were performed to evaluate the pattern and degree of resistance and cross-resistance of several nucleoside analogues against a pair of AZT-resistant and sensitive strains of HIV-1 in human peripheral blood mononuclear (PBM) cells. AZT-resistant and sensitive viruses strain 9F (G910-6) and 10 (H112-2) were obtained through the NIH AIDS Research and Reference Program and propagated in PBM cells. The results on a selection of compounds evaluated at the same multiplicity of infection are shown below and are compared to the prototype LAV strain of HIV-1. The virus yield was determined by a reverse transcriptase assay on disrupted virus that was obtained from supernatant of cells exposed to the compounds for 6 days.

Compound	*EC_{50} (μM) in PBM cells for strain:*			*Fold increase:*
	LAV	*10 (H112-2)*	*9F (G910-6)*	*10/9F*
AZT	0.0015	0.0018	1.15	639
AzddU (CS-87)	0.18	0.08	14.5	181
BCH-189	0.022	0.040	0.24	6
± Dioxolane-T (CS-379)	0.3	0.20	4.93	24

Isolate 9F was clearly resistant to AZT and AzddU in PBM cells, as was previously reported by Larder et al. (AAC **34**:436-441, 1990) in HeLa-CD4+ (HT4-6C) cells. Other compounds evaluated were more inhibitory to the AZT-pretreatment (strain 10) than the post-treatment isolate (9F); the degree of cross-resistance to the AZT-resistant isolate 9F was not as great as that found with AZT or AzddU. These studies performed in primary human lymphocytes could provide the basis for selecting alternate drug treatment for individuals who develop AZT-resistance and also for the development of a rational approach for combined therapeutic regimens in humans. (Supported by the NIH and the VA)

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RESISTANCE OF HCMV TO DHPG, HPMPC AND HPMPA MAPPED TO THE VIRAL DNA POLYMERASE GENE. V. Sullivan[1], K. Biron[2], M. Davis[2], S. Stanat[2], C. Talarico[2], D. Coen[1]. [1]Harvard Medical School, Boston, MA; [2]Wellcome Research Laboratories, Research Triangle Park, N.C

DHPG (ganciclovir) is an antiviral drug used clinically to treat and prevent Human Cytomegalovirus (HCMV) infections, while HPMPA and HPMPC are promising antiviral agents currently under development. We have previously reported the isolation of a DHPG resistant HCMV mutant, 759^{r}D100. Resistance of this mutant was associated with its inability to induce phosphorylation of DHPG in infected cells. However, we have found subsequently that 759^{r}D100 displays resistance to HPMPA, HPMPC, and to 2'-nor-cyclicGMP, antiviral drugs whose anti-herpesvirus action is not dependent on virus-induced phosphorylation. Using marker transfer analysis we have determined that the DHPG resistance of 759^{r}D100 is conferred by two separable mutations; one associated with decreased phosphorylation of DHPG and a second conferring DHPG resistance in a manner unrelated to DHPG phosphorylation. Moreover, we have fine-mapped a mutation conferring part of the $DHPG^{r}$ and all of the $HPMPA^{r}$ and $HPMPC^{r}$ phenotypes of 759^{r}D100 to the DNA polymerase gene. Sequencing studies of 759^{r}D100 have revealed an amino acid alteration in a highly conserved region of the DNA polymerase. These results demonstrate that the HCMV DNA polymerase is a target for the selective action of DHPG, HPMPA and HPMPC. We are currently carrying out further experiments to map the mutations affecting DHPG phosphorylation.