35

Selective Activity of Triciribine Against HIV-1 in Acutely Infected CEM-T4 Cells. M. R. Nassiri, D. E. Ickes, M. Lew, C. Shipman, Jr., L. B. Townsend, and J. C. Drach. University of Michigan, Ann Arbor, Michigan 48109, USA.

The novel tricyclic nucleoside, triciribine (TCN) and its 5'-monophosphate (TCN-P) have potent and selective activity against HIV-1 and HIV-2 in selected cell lines (*AIDS Research and Human Retroviruses,* **9**: 307, 1993). We have extended this work and more rigorously compared the activity of TCN and TCN-P against HIV to cytotoxicity in the same cell line. In a p24 viral antigen assay using CEM-T4 cells acutely infected with HIV-1, IC_{50}'s of 0.013, 0.012 and 0.023 μM were obtained for TCN, TCN-P, and AZT respectively. To examine the cytotoxicity in uninfected cells, drug effects on the cell cycle distribution of CEM-T4 cells were determined by flow cytometry. Cells were treated with 1 to 100 μM of TCN and TCN-P for 12 to 72 hours, stained with propidium iodide, and prepared for flow cytometry. Only minor cell cycle perturbations were caused by either compound at times up to 48 hr. By 72 hr, an effect was more apparent. TCN at 10 and 100 μM, respectively, caused a 13% and 28% accumulation of cells in G_0/G_1 phase; an increase of 27% and 34% was caused by similar concentrations of TCN-P. The delay in cell cycle progression was confirmed by measuring the population doubling time which increased from 20 hr for untreated cultures to 59 and 160 hr with 100 μM of TCN or TCN-P, respectively. Incorporation of [^{32}P]Pi into CEM-T4 cells treated with 100 μM TCN or TCN-P for 48 hr inhibited nucleic acids synthesis by 27% and 45% respectively. Hydrolysis of samples with NaOH revealed only a slight inhibition of DNA synthesis thereby indicating most of the effect was on RNA synthesis. The modest inhibition noted at these high concentrations is markedly different from the cytotoxicity produced in murine L1210 cells at low concentrations (*Cancer Research,* **45**: 6355, 1985). Taken together these new data confirm that the effects of TCN and TCN-P on HIV are well separated from cytotoxic effects in the same human lymphoblastoid cell line thereby providing significant selectivity against the virus. This study was supported by grants U01-AI25739 and R01-AI33332 from the Division of AIDS, National Institute of Allergy and Infectious Diseases.

36

Antiretroviral activity of N-α-acetyl-nona-D arginine amide acetate (ALX40-4C). B. Conway, O. Mpanju, J. Sahai, M. Twist, M. Sumner-Smith. University of Ottawa AIDS Research Group, Ottawa and Allelix Biopharmaceuticals Inc, Mississauga, Ontario, CANADA.

ALX40-4C is a novel peptide, consisting of nine D-arginines. *In vitro*, it competitively inhibits the Tat/TAR interaction, a required step in HIV transactivation. In preliminary studies, laboratory viral strains grown in cell lines have been found to be susceptible to inhibition by ALX40-4C. We sought to extend these observations of antiviral activity, prior to the initiation of clinical trials in Canada. Aliquots of $2x10^6$ PHA-stimulated seronegative mononuclear cells were treated with 0.1-20μM ALX40-4C or 1μM AZT for 24 hours, then infected with HTLV-IIIB (MOI=0.143). Cultures were fed on days 3, 7 and 10, maintaining cells at $1x10^6$/ml. Quantitation of HIV p24 antigen (ng/ml/10^6 cells) in culture supernatants on days 7 and 10 are shown below:

			ALX 40-4C			
	Control	1μM AZT	0.1μM	1μM	5μM	20μM
Day 7	650	0.8	672	362	38	2
Day 10	1030	0.8	257	547	65	1

As expected, this viral strain is exquisitely sensitive to AZT. It is also sensitive to ALX40-4C, with IC50 1.26, 1.46μM and IC90 4.66, 4.68μM, based on day 7 and 10 p24 values respectively. Minimal cytotoxicity was observed up to 20μM, which was approximately as effective as AZT. This drug is effective in mononuclear cell models. *In vitro* evaluation of clinical viral strains as well as *in vivo* phase I/II pharmacokinetic and 28 day tolerance/efficacy studies are planned in our centre.