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Intracellular activation and cytotoxicity of combinations of 3'-azido-3'-deoxythymidine and 2',3'-dideoxyinosine. S. Palmer and S. Cox. Virology Dept, Swedish Institute for Infectious Disease Control, Karolinska Institute, 10521 Stockholm, Sweden.

The combination of azidothymidine (AZT) and dideoxyinosine (ddI) exhibits in vitro synergistic inhibition of human immunodeficiency virus (HIV). An important consideration for clinical use of AZT and ddI in combination is that of the choice of the ratio between these two drugs and the cytotoxicity of the combination. We therefore measured the intracellular activation and cyto -toxicity of AZT and ddI when combined at three different clinically relevant combinations of 1:1, 1:10 and 10:1. To assess the intracellular activation, we incubated CEM lymphocytes with three combinations of AZT and ddI. The levels of AZT TP and ddA TP formed for each combination were then quantified by ion exchange HPLC. We found the activation of ddI to ddA TP was increased two-fold in all three combinations with AZT, compared to ddI alone. Increasing the concentration of AZT in the combinaitons did not give further increases in the activation of ddI and only produced a slight increase in the amount of AZT TP formed. We examined the cytotoxicity of the 1:1, 1:10 and 10:1 combinations of AZT and ddI in CEM cells, PHA-stimulated and resting PBMCs. CEM cells were the least sensitive overall to the drugs. AZT showed greater cytotoxicity in stimulated PBMCs than resting PBMCs whereas the reverse was true for ddI. This could be explained by the different activation pathways of these two drugs. The 1:1 and 10:1 AZT:ddI combinations showed reduced toxicity compared to the separate drugs. We conclude that the increased activation of ddI when combined with AZT at ratios of 1:1, 1:10 or 10:1 may explain the synergy exhibited by the combination of these drugs against HIV. The reduced cytotoxicity of the 1:1 and 10:1 combinations to -gether with the increased activation of ddI makes them a promising choice.

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Activity of AZT and ddI When Used in Combination With Other Mechanistically Diverse Inhibitors of HIV Replication. Robert W. Buckheit, Jr. and Julie D. Russell, Southern Research Institute-Frederick Research Center, Frederick, MD USA

We have evaluated the activity of AZT and ddI in combination with anti-HIV agents representative of each of many mechanistic classes, including various inhibitors of attachment and fusion, reverse transcription, protease, glycosylation, and trans-activation. The activity of the agents in combination with the nucleosides AZT and ddI was uniform within each of the distinct mechanistic classes. The combination of AZT or ddI with inhibitors of virus attachment or fusion (dextran sulfate, Chicago Sky Blue, ISIS 5320) yielded additive anti-HIV activity. At high concentration of the sulfated polysaccharide or sulfonated dye, the activity became antagonistic. In combination with other nucleoside RT inhibitors (ddC, carbovir) synergistic anti-HIV activity was detected. Significant synergy was also detected when AZT or ddI were used in combination with nonnucleoside RT inhibitors (TIBO, thiazolobenzimidazole, UC38, calanolide A) or phosphonoformic acid. No antagonism was detected with these combinations of reverse transcriptase inhibitors. AZT used in combination with inhibitors of HIV protease routinely yielded synergistic anti-HIV activity, without evidence of antagonism. Inhibitors of glycosylation and tat yielded additive to slightly synergistic anti-HIV activity when used in combination with AZT and ddI. Antagonistic action was observed with the combination of AZT with ribavirin and mixed synergy and antagonism was observed with ddI and ribavirin. Our data were evaluated using the Prichard and Shipman three dimensional model. We have also evaluated these data using the Chou and Talalay model. Similar interpretation of the results was obtained with both models. Finally, we have evaluated the interaction of three compounds in the inhibition of HIV replication. The use of two NNRTIs with AZT, or a single NNRTI with both AZT and ddC have been evaluated as potential therapeutic strategies. The results of our ongoing studies to evaluate the activity of the three drug combination will be presented.