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Pharmacokinetics of β-D-2',3'-Didehydro-2',3'-dideoxy-5-fluorocytidine (D-D4FC) in Rhesus Monkeys.

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β-D-2',3'-Didehydro-2',3'-dideoxy-5-fluorocytidine (D-D4FC) has potent and selective anti-HIV and HBV activity *in vitro*. The single-dose pharmacokinetic parameters of D-D4FC in rhesus monkeys after intravenous and oral administration of 33.3 mg/kg were determined using a two compartment model. Due to the lability of D-D4FC in acid, $NaHCO_3$ buffer solution was used for oral administration. The average value for the terminal half-life, $t_{1/2\beta}$, was 3.2 hr (CV = 7.4%). Average values for renal clearance (Cl_{renal}) and for total systemic clearance (Cl_{sys}) were 0.31 (CV = 13.4%) and 0.41 (CV = 4.7%) $l \cdot kg^{-1} \cdot hr^{-1}$, respectively. Oral bioavailability of D-D4FC was incomplete, with an average of 47.6% (CV = 32.5%) of the dose reaching the systemic circulation. More than 74% of the compound was recovered in the urine in 8 hr, indicating that D-D4FC was eliminated mainly by renal excretion. D-D4FC was detected in CSF at similar concentrations for both i.v. and oral routes. The favorable pharmacological profile of D-D4FC warrants its development as an antiviral agent.

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In Vivo Activity Of An Antisense Oligonucleotide Targeted Against The E1 Region Of Human Papillomavirus

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We have studied the inhibitory activity of antisense oligodeoxyribonucleotides (ODNs) that target expression of the E1 helicase region of human papillomavirus (HPV) types 6 and 11. Activity of antisense ODNs *in vitro* was measured using mammalian cells transfected with an E1-luciferase reporter gene. Selected compounds were further examined *in vivo* using a kidney xenograft model in which HPV-infected foreskin fragments are implanted under the kidney capsule of a nude mouse. A phosphorothioate 2'-O-methyl RNA hybrid (changes at the 5' end) ODN targeting the E1 AUG of HPV (known as HPV1 0x5 OMe) had good inhibitory activity in the xenograft model and reduced HPV induced condyloma growth by 95%. A mismatched ODN in which the some of the guanine bases were replaced by adenine was inactive in this model. Further studies of the activity of these ODNs was made using the mCMV infection model in mice. HPV1 0x5 OMe, but not the mismatched ODN, surprisingly protected the mice from the lethal effects of mCMV. To determine the mechanism of action of HPV1 0x5 OMe we tested this compound against another human papilloma virus - HPV40 - which did not have the same sense sequence as HPV6 and 11, and was therefore mismatched to the compound. HPV1 0x5 OMe inhibited condyloma growth induced by both HPV11 and HPV40 to the same degree. We believe that the action of HPV1 0x5 OMe in the xenograft model is not totally related to an antisense mechanism, one possible mechanism is via an immune stimulatory effect.

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Detection of Soluble Nuclear Matrix Protein (NMP) Released from Apoptotic Nuclei of Human Papillomavirus (HPV) Positive Cell Lines Treated with Cidofovir

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Acyclic nucleoside phosphonates (ANPs), in particular cidofovir, inhibit the growth of rapidly proliferating cells. This phenomenon is mainly observed in HPV-, adenovirus- and SV40-transformed cell lines. We have shown by a cellular DNA fragmentation ELISA and by cell cycle analysis that the process leading to cell death following treatment with ANPs is due to apoptosis (programmed cell death). Apoptosis is associated with changes in several cellular processes. During apoptosis, dense chromatin masses increase in number until the nucleus becomes pyknotic. The skeletal structure for the topological organization of chromatin in the interphase nucleus is NMP, and breakdown of the overall nuclear structure results in the disruption of normal chromosomal nuclear matrix interactions. We have used a sensitive NMP ELISA that demonstrates the release of NMP in a soluble form from the apoptotic nucleus. A direct relationship between the amount of detectable soluble NMP and the number of dead cells was observed when CK-1 cells (HPV-33$^+$) and HeLa cells (HPV-18$^+$) were treated with different concentrations of cidofovir and cytarabine (AraC). Thus, appearance of detectable soluble NMP, one of the products of the segmentation of the nucleus, confirms that the process leading to cell death following treatment with cidofovir is apoptosis.

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A Double-Blind, Placebo-Controlled Study of Cidofovir Gel for Human Papillomavirus (HPV)-Associated Genital Warts. R. SNOECK,[1*] M. BOSSENS,[2] D. PARENT,[3] B. DELAERE,[4] H. DE GREEF,[5] E. DE CLERCQ,[1] S. SAFRIN,[6] B. McGUIRE,[6] H. S. JAFFE.[6] [1]Rega Institute, Leuven Belgium; [2]Association Hospitalière Etterbeek-Ixelles, Bruxelles; [3]Erasme Hospital, Bruxelles; [4]St. Luc Hospital, Bruxelles; [5]U.Z. St. Rafaël, Leuven; [6]Gilead Sciences, Inc., Foster City CA.

Cidofovir is an acyclic nucleotide analog with anti-HPV activity in animal models and anti-proliferative effect in HPV-infected cell lines. Thirty-one immuncompetent patients (18 women, 13 men) with biopsy-proven HPV-associated anogenital warts (untreated, refractory, or recurrent) were stratified by total wart area and randomized to receive 1% cidofovir (CDV) or placebo gel in a 2:1 ratio, applied qd x 5 days every other week for a maximum of 6 cycles (12 wks). Wart response at week 12 was categorized as: complete (100% clearance), partial (≥50% area decrease), no change (25-50% decrease), and progression (>25% increase). Previous therapy for warts included podofilox (37%), cryotherapy/electrocautery (34%), laser (20%), other (excision, 5-FU;17%); 57% of pts had no prior treatment. Baseline median wart surface area was 56 mm^2 (range,8-1756 mm^2); median wart number was 8.5 (range, 1-20). Nine of 19 CDV pts (47%) had complete clearance vs. none of 11 (0%) placebo pts (p=.006). Sixteen CDV pts (80%) had a complete or partial response vs. 2 (18%) placebo pts (p=.001). Only 1 of 8 complete responders followed to date (median follow-up, 180 days; range, 127-252 days) had recurrence of same-site warts. Reversible application site reactions (pain, pruritus, ulceration, rash) occurred in 65% CDV vs. 55% placebo pts (p=0.7). No evidence of systemic toxicity has been seen.