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Pharmacokinetics of 3'-fluoro-guanosine (FLG) in cynomolgus monkeys

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Recently FLG has been shown to be active against duck hepatitis B virus in ducks and is therefore a compound of potential value in the treatment of the corresponding disease in humans. Here, the basic pharmacokinetic properties of FLG in a non-human primate species, the cynomolgus monkey, is described. In 4 monkeys FLG 3 mg/kg was given iv. In 3 of these 10 mg/kg was given po at another occasion at least one week apart from the iv administration. HPLC with UV-detection was used to analyse the drug. Protein binding to human plasma was determined by microdialysis. The results are given as mean±SEM. After iv administration the half-life was 27±3 min, the clearance was 38±9 ml/min/kg and the volume of distribution was 1.34±0.13 l/kg. No more than 5% of the dose was recovered as unchanged FLG in the urine. No glucuronide of FLG could be demonstrated by β-glucuronidase treatment of the urine. Protein binding was not detectable. The oral availability was small but similar in the 3 monkeys studied, 4±1%, with C_{max} 0.27±0.03 μM. The pKa of FLG was 9.25±0.06 and the octanol/water (pH 7.4) partition was 0.193±0.006. Thus, the pharmacokinetics of FLG is similar to that of other guanosine analogues.

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COMPETITIVE ELISA FOR D4T : APPLICATION TO CLINICAL PHARMACOLOGY.

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I) Purpose of the study ::D4T, a thymidine analog with potent anti HIV activity *in vitro*, is currently investigated as therapy for patients with advanced HIV infection. D4T is a prodrug which is converted by cellular thymidine kinases to the active metabolite D4T triphosphate (D4T-TP), acting as competitive inhibitor of the HIV encoded reverse transcriptase. Because dose-limiting toxicity is associated with D4T therapy and level of intracellular D4T-TP is dependent on various factors such as the cell type, the activation state or the presence of other antiviral agents, measurement of D4T and its phosphate forms is essential.

II) Methods :

In the present study, using anti-D4T rabbit antibodies raised against D4T hemisuccinate-bovine serum albumin conjugate, a D4T hemisuccinate-peroxidase as tracer and a microtiter plate coated with anti-rabbit IgG as separator system, a one-step convenient competitive ELISA method was developed for assaying D4T.

III) Summary of results :

The method was capable to specifically detect 2 ng/ml of D4T in ultrafiltrates from plasmas previously separated on microconcentrator devices. Thymidine, azido thymidine and D4T monophosphate which closely resembles D4T, cross-reacted at 0,04%, 0,25% and 1,8% respectively. The method was applied for quantitating intracellular D4T in CEM and Molt 4 cell lines and monitoring plasmatic D4T in patients with advanced HIV infection. Additionnally, this technique was extended to the indirect measurement of intracellular phosphorylated D4T metabolites following *reverse phase* separation of cell extracts and treatment of fractions with alkaline phosphatase.

IV) Conclusions :

The extrapolation of this method to another drugs and their metabolites, such as AZT, DDI, DDC, AZT-TP, DDI-TP, DDC-TP..., will be investigated.