43

TYPE-1 PLASMINOGEN ACTIVATOR INHIBITOR (PAI-1): CELL-SPECIFIC EXPRESSION AND HORMONAL REGULATION IN HUMAN CELL LINES

L.R. Lund(1), A. Riccio(2), M. Mayer(1,3), B. Georg(1,3), K. Danø(1), F. Blasi(2) and P.A. Andreasen(1,3)
(1)Finsen Laboratory, Rigshospitalet, Copenhagen, (2)Institute of Microbiology, University of Copenhagen, (3)Institute of Biochemistry C, University of Copenhagen, Denmark.

We have screened 15 human, normal and neoplastic cell lines for PAI-1 production in the absence and the presence of glucocorticoids by the use of an enzyne-lniked immunosorbent assay. The basal PAI-1 production varied approximately 5000-fold between the cell lines; only 3 cell lines responded to glucocorticoids by an increased PAI-1 production. Northern- and dot blot analyses with the glucocorticoid responsive cell line HT-1080 showed that the increased production of PAI-1 following glucocorticoid treatment was caused by an increased level of PAI-1 mRNA. Nuclear transcription assays showed that the increase was at least due to a partly increase in the rate of transcription of the PAI-1 gene. Transfection studies with a 874 basepair fragment of the 5' flanking region of the PAI-1 gene fused to the coding sequence of the E.coli CAT gene showed that the flanking region has promoter activity. The same fragment contains information enough to confer glucocorticoid responsiveness to the fusion gene transfected into HT-1080 cells.

44

FURA-2 MEASUREMENT OF CYTOSOLIC FREE Ca2+, (Ca2+)i, IN DAUNORUBICIN (DNR) AND VINCRISTINE (VCR) RESISTANT EHRLICH ASCITES TUMOR CELLS.

P. Bouchelouche[1], T. Skovsgaard[2], M.Sehested[3] and E. Friche[2].
[1]Dept. of Clinical Chemistry and [3]Pathology, Herlev university Hospital, [2]Dept. of Internal Medicine, The Finsen Institute, Copenhagen, DK.

Calcium channel blockers, such as verapamil, are capable of reversing acquired cellular resistance to DNR and VCR presumably by increasing drug uptake in resistant cells. However, the role of (Ca2+)i per se in this process is uncertain. The aim of the present study was to investigate the relation between calcium, verapamil and drug accumulation.
(Ca2+)i was measured by the new fluorescent calcium indicator Fura-2. Fluorescence of the Fura-2 loaded cells was measured using excitation wavelength 340 nm and emission wavelength 510 nm. Wild-type tumor cells (EHR2) had significantly higher resting levels of (Ca2+)i (140-180 nM), than EHR2/DNR+ and EHR2/VCR+ resistant sublines (50-80 nM), when the extracellular medium contained calcium. However, in calcium free medium no difference was found. No change in DNR accumulation was seen neither when calcium was absent from the extracellular medium, nor when the cells were calcium depleted, nor when intracellular calcium was chelated. Verapamil also remained active in the absence of extra-/intracellular calcium. Therefore, we conclude that
1)resistant cells have a higher Ca2+ efflux rate across the plasma membrane and
2)calcium is not involved in the action of verapamil on drug accumulation in resistant cells.

45

THE BIOAVAILABILITY OF METOPIMAZIN (VOGALENE^{R}): A PHARMACOKINETIC STUDY.

Herrstedt J., Jørgensen M. & Angelo H.
Bispebjerg Hospital, Copenhagen & Copenhagen University Hospital, Herlev.

Metopimazin (MPZ) in doses below the toxic level has demonstrated a certain effect compared to placebo in the treatment of emesis and vomiting induced by cytostatic agents. Aiming at determining the relevant dose level and dose interval we have investigated the bioavailability of MPZ.
Six healthy volunteers participated in four subtrials (a-d) given MPZ as a single dose (oral): a) 20 mg preprandially b) 50 mg preprandially c) 20 mg postprandially and d) 5o mg postprandially. Blood samples were drawn regularly in the period 0-8 hours. The serum concentration of MPZ and the acid metabolite (AMPZ) were determined by the HPLC method. Time/concentration curves were established. The absorption showed significant interperson variation expressed by AUC and C-max. Food decreased the bioavailability of MPZ. MPZ showed no saturation kinetics at the dose level 20-50 mg. Only minor side effects were observed.
We suggest that MPZ is dosed individually and is taken preprandially.

46

ULTRASONICALLY DETECTION OF AXILLARY METASTASES IN PATIENTS TREATED OF BREAST CANCER.

M.Jørgensen, K.D.Pedersen, K.E.Fredfeldt, A. Glenthøj and S.A.Engelholm. Dept. of Oncology, Ultrasound and Pathology, Copenhagen University Hospital, Herlev.

Two prospective studies were made to outline the diagnostic value of ultrasound (US) in patients (pts) treated of breast cancer.
1) 60 pts with regional symptoms, but normal clinical examination underwent US examination with a 5-7 MHz small part scanner in real time. If focal processes were visualized US guided fine needle biopsy (UFNB) was done.
2) 53 pts with suspect palpation all had palpation guided fine needle biopsy UPFNB) done, and then US examination and if focal processes were visualized, also UFNB. In "1)" UFNB detected malignancy (M) in 7(12)%. US were normal or with minor changes (e.g. edema, fibrosis) in 46(77)%. In "2)" M was found in 31 pts. Of these UFNB detected M in 30, in one US was pathological, but UFNB was not feasible. PFNB detected M in 26 pts. Of 22 pts., where M was not found, 11 had this verified by surgical biopsy; another 10 had no signs of M with a follow-up of in mean 16 mths (13-24), and only one pt had locoregional recurrence after 6 mths. Interestingly US was without focal processes, in 14 pts with histological benign, palpable nodes. We concluded: US can detect non-palpable axillary metastases. In some cases US could disprove suspected M. US and UFNB can give additional information if PFNB is inconclusive.