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Study of estradiol receptor (ER) has been used as reliable tool for predicting results of hormonal treatment in human breast cancer. Approximately 40% of receptor positive patients did not respond favourably. In our study we used estradiol induced peroxidase (EC I.II.I.7) as another marker of hormone dependent breast cancer. In 96 primary breast carcinomas we determined ER and peroxidase activity after 24 hr stimulation in organ culture with 10^{-8}M estradiol. Peroxidase assays were performed by the rate of oxidation of guaiacol and were expressed as unit per mg protein (over 1 U/100 mg protein as positive). 33 carcinomas were ER and peroxidase positive and 32 of them responded favourably after endocrine treatment; 60 carcinomas were ER and peroxidase negative and only 3 of them responded. Estradiol induced peroxidase is therefore a potentially useful marker of hormone dependent human breast carinomas.

METASTATIC GROWTH OF HEPATOCARCINOMA CELLS IN F344 RATS AFTER SUBRENAL CAPSULE TRANSPLANTATION OF PRIMARY TUMOURS

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A metastasis-forming hepatocellular carcinoma was induced in a F344 male rat by a single injection of MNU of newborn age. The tumour was maintained by serial passage in F344 rats and metastases were found without exception on the peritoneum and in the parathymical lymph nodes. A new method was elaborated for standardization of tumour growth: transplant discs with equal size were prepared and put under the left kidney capsule. The progression of primary tumour tissue and its metastases could be followed by protein and DNA determinations. Enhanced tumour metastasis was observed after ablation of the left kidney even three days after tumour-transplantation. It is proposed that metastasis formation is a very early phenomenon, but metastatic cell growth is suppressed by primary tumour cells.

BENZO(A)PYRENE-DNA ADDUCTS - IMPLICATIONS OF EXPERIMENTAL AND HUMAN DATA FOR MONITORING

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Benzo(a)pyrene (BP) is an animal carcinogen, and it may contribute to lung cancer caused by cigarette smoke and occupational exposure to polycyclic aromatic hydrocarbons (PAH). We are using synchronous fluorescence spectrophotometry (SFS) and ultrasensitive enzymatic radioimmunoassay (USERIA) to measure BPDE-DNA-adducts, the putative carcinogenic lesion caused by BP-exposure. Although in controlled experimental situations, e.g. in animals treated in vivo with BP, both methods are very sensitive, quantitative and correlate well with each other, there are some unanswered questions as to the in vivo monitoring. The specificity of the methods is not complete when isolated PAH-DNA adducts are studied, but may still be adequate for in vivo monitoring. We have found some positive cases among human DNA isolated from blood cells of work-exposed or from placenta from smoking mothers. To further evaluate the contribution of cigarette smoking and the application of the methods we use, we are trying to set up an animal model. Although we have found a dose-dependent increase in the BPDE-DNA adduct formation in several organs after in vivo treatment of C57BL/6 and DBA/2 mice with BP, no adducts have been detected after cigarette smoke exposure in vivo, or injection of cigarette smoke condensate (CSC) or neutral fraction of CSC, even in the case where the AHH-activity was induced. It seems that in human tissues, similarly, there is a far-from-perfect correlation between the AHH-activity and in vivo or in vitro formed adducts.

MONO-OXYGENASE CATALYZED REACTIONS AND BINDING OF BENZO(A)PYRENE TO DNA IN HUMAN TISSUES. ROLE IN SUSCEPTIBILITY TO CHEMICALLY INDUCED CANCER

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We have studied whether the in vitro measured monooxygenase (MO) activities in human tissues are associated with susceptibility to chemical-induced cancers. Although activity and inducibility of various MOs show large inter-organ and inter-individual differences the induction of aryl hydrocarbon (benzo(a)pyrene) hydroxylase (AHH) seems to be at least partially "systemically" regulated, thus

supporting the notion, that lymphocytes could be used as a model for primary target tissue, e.g. lungs. However, we have not seen any significant differences in lymphocyte AHH activity and inducibility between lung cancer patients and controls.

We have also studied whether MO activities predict the in vivo or in vitro formation of benzo(a)pyrene diolepoxide (BPDE)-DNA adducts, the model lesion for PAH-induced carcinogenesis. We have shown that the nature of P450 isozyme is of importance for the activation ability, whereas the magnitude of activity seems to be of lesser importance. This was demonstrated with the aid of monoclonal antibodies (MAb) to different P450 isozymes. For example, the MAb to rat MC-induced liver P-450 inhibited AHH in placenta, but not in liver. It readily inhibited the in vitro formation of BPDE-DNA adducts in placenta, but not in liver. The MAb to phenobarbital-induced isozyme did not have these effects.

THE EFFECT OF INHIBITION OF MITOCHONDRIAL PROTEIN SYNTHESIS ON THE GROWTH KINETICS OF A RAT LEUKAEMIA

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Mitochondria (mt) contain DNA coding for several subunits of components of the oxidative phosphorylation system and a specific system to transcribe and translate this DNA. Inhibition of the expression of mt-genes finally diminishes the capacity for oxidative phosphorylation to an extent that cell functions (e.g. proliferation) become impaired. This has been demonstrated already in several tumour systems. Specific and continuous impairment of mt-protein synthesis by treatment with tetracyclines results also in growth inhibition of a leukaemia in the rat - it even leads to its disappearance. Cytostasis is achieved more rapidly and the rat of cytolysis is faster when tetracycline treatment is started in later stages of tumour progression. Our studies indicate that this is due to interference of tetracyclines with the cytostatic and cytolytic effects of corticosteroids on the growth of this tumour. As tetracycline treatment has lasted longer, the anti-tumour effects of (endogeneous) corticosteroids become less. It is suggested that mt-protein synthesis is required for the action of corticosteroids on leukaemic cells.

PROVIRUS INTEGRATION IN [90]Sr -INDUCED OSTEOSARCOMAS OF CFF1 MICE

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The development of [224]Ra and [90]Sr induced osteosarcomas in mice is accompanied by the expression of endogenous retroviruses in bone tissues of the treated animals in the early latency period, and later in the osteosarcomas. Using the Southern blotting procedure, we have studied the presence of somatically acquired proviruses in genomic DNA isolated from seven primary [90]Sr induced osteosarcomas and one osteosarcoma cell line (0-127a1) of the CF1 mouse strain. Newly integrated ecotropic proviruses were detected with specific hybridization probes in four primary tumours. In contrast, genomic DNA from cultured osteosarcoma cells harboured additional ecotropic recombinant (MCF-related) proviruses. No integrations were found in the vicinity (22 kbp) of c-myc. The c-myc locus is amplified in two out of eight tumour DNAs. According to our data, detectable integrations of activated retroviruses do not appear to be an essential requisite for the development of radiogenic osteosarcomas in mice, but in some cases, clonal or oligoclonal integrations might have been responsible for the deregulation of a nearby putative oncogene, allowing cells to escape normal growth control in vivo.

REVERTANTS OF METHIONINE-DEPENDENT H-ras-1 ONCOGENE-TRANSFORMED CELLS

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Methionine-dependence is a metabolic defect reported to be exhibited by many transformed and malignant human or animal cells (Mechan et al, BBRC, 117: 429, 1983). This defect is characterized by the inability of cells in culture to grow in a medium where methionine has been replaced by its immediate metabolic precursor, homocysteine. The biochemical basis of this phenomenon is not understood. We have shown recently that the activated H-ras-1 oncogene, derived from the EJ human carcinoma line, induces methionine-requirement after transfection in