anti-cancer drugs which inhibit DNA synthesis can increase the frequency of gene amplification. The currently favoured model for the mechanism of gene amplification is that of saltatory replication whereby unscheduled DNA replication creates strands of DNA that are not attached to the chromosome; such re-replicated DNA may be observed cytologically as double minutes (DMs) or homogeneously staining regions (HSRs).

We have investigated whether the anti-cancer drug hydroxyurea can induce this mechanism of gene amplification in human neuroblastoma CHP-100 cells. DNA double labelling techniques have revealed no evidence of re-replication of DNA following hydroxyurea treatment. Therefore, it is unlikely that hydroxyurea can induce gene amplification by this mechanism.

This work was funded by the Yorkshire Cancer Research Campaign.

THIOL STATUS OF NORMAL HUMAN BRONCHIAL EPITHELIAL CELLS AND FIBROBLASTS

L.Atzori, K.Sundqvist, J.M.Dypbukt, I.Cotgreave, P.Moldéus and R.C.Grafström

Department of Toxicology, Karolinska Institutet, Stockholm, Sweden.

The content of total sulphydryls (SH) and low molecular weight thiols (LMWT) including reduced glutathione (GSH), and cysteine, oxidized glutathione (GSSG), cystine and mixed protein disulphides as determined in human bronchial epithelial cells and fibroblasts. Epithelial cells had signficantly higher levels of total SH than fibroblasts, 75 as compared to 53 nmol of SH per 10^6 cells, respectively. In both types of cells, qualitative analysis indicated similar proportions among the various LMWT where GSH was found to be the major thiol. For both cell types, passage in culture caused an immediate decrease in total thiols and also changed ratios among different LMWT. Continued culture caused a marked peak in GSH synthesis which preceded cellular proliferation. Furthermore, the proportion of GSSG plus mixed disulphides was significantly higher before the cells entered the growth phase. During logarithmic growth, the amount of GSH was markedly decreased. Prolonged maintenance of fibroblasts at confluence, did not cause further change in SH. The results indicate variations in SH content between different human cell types and implicate the importance of LMWT in growth regulation.

DNA MEASUREMENTS FOR EFFECTIVE CHEMOTHERAPY OF HÜRTHLE CELL CARCINOMA

M.Auersperg(1), R.Zorc(2), J.Petric(1), M.Us-Krašovec(1) and O.Porenta(2)

(1)The Institute of Oncology and (2)The Institute of Histology and Embriology, Ljubljana, Yugoslavia

Inoperable or disseminated Hürthle cell carcinoma is a therapeutic challenge as therapy with [131]I or radiation or chemotherapy is usually ineffective. In order to find an effective chemotherapy, the influence of vinblastine (VLB) (2 mg bolus or infusion over 6, 12, 24 hr) was studied in 5 patients (4 women, 1 man, aged 43 to 69 yrs). Four patients had distant metastases, one locoregional disease only. Thin-needle aspiration biopsies of tumours (1 primary, 4 metastases) were performed before and repeatedly after VLB applications. The smears were stained after Feulgen and were used for cytophotometric DNA measurements. VLB produced an increase of cells in S phase compartment. On the basis of changes produced in the DNA distribution pattern by the test dose of VLB, chemotherapy was planned: either a sequence of 3 VLB infusions with individual intervals or a combination of VLB, cisplatinum, methostrexate, bleomycin or adriamycin was used. All 5 patients responded - 1 CR, 4 PR. Chemotherapy was combined with surgery in 1 and radiation in 2 patients. Two out of 5 patients show no evidence of disease 3.1 years after therapy, 2 continue chemotherapy, 1 patient is dead of other causes.

DETECTION OF ANTIBODIES AGAINST AFLATOXIN-CONJUGATE IN SERA FROM AFRICAN AND DANISH POPULATIONS

H.Autrup and T.Seremet

Laboratory of Environmental Carcinogenesis, The Fibiger Institute, Copenhagen, Denmark.

A sensitive ELISA assay has been developed to detect antibody activity against aflatoxin (AFB) in human sera. Antibodies to an epitope on AFB-BSA were detected in all sera collected in Kenya. The specific activity showed a trimodal distribution. The high activity group had a higher frequency of recent AFB exposure, as measured by urinary excretion of aflatoxin-guanine, than the low activity group. Little or no activity was detected in Danish sera. Animal experiments indicate that the specific activity depends on the metabolism of AFB to its ultimate carcinogenic form. The activity in rat sera was inhibited in a competitive assay by an aflatoxin-like antigenic material found in

human urine, and to a lesser extent by AFB and its primary metabolites. The results suggest that carcinogen-albumin adducts formed after carcinogen exposure can act as an immunogen.

(Supported by the Danish Cancer Society.)

SUSCEPTIBILITY TO LUNG CANCER

Herman Autrup

Laboratory of Environmental Carcinogenesis, The Fibiger Institute, 70 Ndr Frihavnsgade, DK-2100, Copenhagen Ø, Denmark

Lung cancer is the most prevalent form of cancer in the industrialised world, and smoking of tobacco products is considered the single most important etiological factor. One of the plausible host factors is the genes controlling both the oxidative metabolism and deactivation of the tobacco carcinogens. A wide inter-individual variation in the primary metabolism of tobacco associated carcinogens, e.g. benzo(a)pyrene, in the target tissue has been reported. Glutathione transferases are involved in the secondary metabolism, and the genes expressing the isozyme(s) is another host genetic determinant. The initial damage introduced in cellular DNA by the ultimate form of the tobacco smoke carcinogens can be repaired by DNA repair enzymes. One of these, O-6 alkyltransferase, shows a wide inter-individual variation in human bronchial epithelial cells.

Other acquired or inherited diseases do influence an individual's risk of developing lung cancer, i.e. sarcoidosis and scar (tuberculosis).

THE EFFECT OF PHENOLIC ANTIOXIDANT, OCTYL GALLATE ON THE BINDING OF BENZO(A)PYRENE METABOLITES TO NUCLEAR DNA

W.Baer-Dubowska and J.Gnojkowski

Department of Biochemistry, Academy of Medicine, Poznan, Poland.

The effect of intraperitoneal (ip) administration of octyl gallate on the formation of benzo(a)pyrene (BP) metabolites-DNA adducts in vitro in rat liver, kidney and lung nuclei was investigated. Male Wistar rats received 2 ip injections per week for 2 weeks. Nuclei were incubated with [^{3}H]BP and DNA was purified and enzymically hydrolysed to deoxyribonucleosides. The BP-DNA adducts were resolved by HPLC. In the nuclei of all tissues the nature of adducts was identical but the level of BP binding was different - the highest in the liver and the lowest in the lung nuclei. The major adducts identified were the BP-4,5-oxide-DNA and the BP-trans-7,8-diol-9,10-epoxide deoxyguanosine adduct. Treatment of animals with octyl gallate decreased the total binding level of BP to DNA of the liver nuclei and the formation of all adducts by 40%. In the kidney nuclei the total binding level and formation of adducts were slightly elevated and in the lung nuclei unchanged. These results indicate that octyl gallate may play a certain role in the inhibition of BP-induced carcinogenesis but this effect is tissue specific.

EFFECTS OF ASCORBIC ACID (AA) ON GENOTOXIC ACTIVITY OF CHEMICAL CARCINOGENS

R.Balansky, P.Blagoeva, Z.Mircheva and I.Stoichev

Institute of Oncology, Sofia 1156, Bulgaria

The influence of AA on mutagenic and carcinogenic activity of benzo(a)pyrene (BP), tobacco smoke (TS), urethane (U) and diethylnitrosamine (DEN) were investigated in vitro and in vivo.

AA added to the top agar (0.2 to 2.0mg/plate) suppressed in about 50% the mutagenic activity of BP in S. typhimurium TA98 but not in TA100 and failed to influence the mutagenic effect of TS. AA (0.3%, 1.0%, 1.5%, with the drinking water) depressed the clastogenic activity of BP (2.0mg/mouse) and TS without influencing the effect of U (1.0g/kg). When applied after the U administration but not given together with the carcinogen, AA inhibited the lung carcinogenesis in mice. An inhibition of liver carcinogenesis was also established in rats treated with DEN (80mg/kg) and AA.

COMBINATION TREATMENT OF 4-EPI-DOXORUBICIN AND RADIATION ON HAMSTER LUNG CELLS

Jasna Ban, M.Bistrović and K.Kolarić

Central Institute for Tumors and Allied Diseases, Zagreb, Yugoslavia

The purpose of this study was directed to test the type and the degree of cytotoxic effects of epirubicin and radiation in combined treatment on Chinese hamster lung cells in vitro. Experiments were performed with proliferating tissue culture cells. Cell killing was determined by colony-forming ability. The maximum killing effects were obtained when simultaneous action of drug treatment and irradiation occurred. Their interaction was synergistic. Synergism depended on time of drug incubation (epirubicin present for 1