intestinal diamine oxidase activity, both being associated with the tumour growth.

THE EPIDEMIOLOGY OF ENDOGENOUS NITROSATION IN MAN

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Epidemiological investigations designed to study the role of endogenous N-nitroso compounds in human cancer have produced inconclusive results. The development of the N-nitrosoproline (NPRO) test (1) has made possible the quantitative estimation of endogenous nitrosation. We have used this test to study nitrosating ability in relation to the risk of gastric cancer.

Healthy males aged 20 to 35 years, resident in two regions of Italy with contrasting mortality from gastric cancer have been compared using the test. NPRO is measured in 12-hour urine samples following a dose of 500 mg proline. In a separate study, patient groups with precursor lesions for gastric cancer were compared with those having normal gastric epithelia. In this case 24-hour urine samples were analysed following proline and nitrate doses.

The results of these studies have been evaluated in relation to dietary characteristics of the groups.

(1) Ohshima H. and Bartsch H. Cancer Res. 41: 3658-3662, 1981.

THERAPEUTIC EFFECT OF CHEMOIMMUNOTHERAPY ON LYMPHOMA BEARING MICE

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The therapeutic effect of a combination therapy employing anti-tumour immune lymphocytes and either doxorubicin (DX) or cis-diamminedichloroplatinum II (DDP) was tested on BALB/c mice bearing YC-8, a weakly immunogenic lymphoma. Mice inoculated with 10^4 YC-8 tumour cells given i.v. all died with liver metastasis. Therapy with immune lymphocytes alone ($30x10^6$ i.v. every 2 days x 3) gave an 80% cure rate when started 3 days after tumour inoculation; when treatment was delayed (5 days) only 20% of the mice were cured. Given at 7 days, immune lymphocytes were ineffective. DX (10 mg/kg) and DDP (6 mg/kg) i.p. gave a significant increase in survival time at all days tested but no cures were obtained. DDP was slightly more active than DX. The association of DDP (day 5) with immunotherapy (day 7) was more effective (54% cures) than DDP alone and day 5 or 7 immunotherapy alone. Only a slight increase in life span was found by combining immunotherapy with DX. The results suggest that combination of chemotherapy and immunotherapy may improve the effects of each treatment alone.

BINDING FOR cis-DIAMMINEDICHLOROPLATINUM (II) TO DINUCLEOTIDES

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Cis-diamminedichloroplatinum (II) (cis-Pt) is a widely used anticancer agent, whose main target is thought to be DNA. In this study, we have incubated cis-Pt with four homodinucleotides (GpG, ApA, CpC, and UpU) and six heterodinucleotides (GpC, CpG, GpU, UpG, GpA, and ApG) at pH 6 at 37 °C. The reaction products were purified by HPLC. Cis-Pt reacted equally well with all guanosine-containing dinucleotides, while the reaction with ApA was much slower. With CpC and UpU no reaction products were formed. The most important products were characterised by ^{1}H NMR spectra. In all the heterodinucleotides except the ones containing uridine, the main Pt-adduct was an intramolecular cross-link, in which the other binding site of cis-Pt was the N-7 atom of guanosine. The other products were intermolecular cross-links and monofunctional Pt-adducts. In the case of homodinucleotides GpG gave almost entirely intramolecular cross-links, and ApA gave both monofunctional and bifunctional Pt-adducts. These results suggest that in DNA cis-Pt is first bound to the N-7 atom of guanine, and then to another base, which may be either guanine or adenine, or even cytosine or thymine.

ISOLATION OF LYMPHOCYTE CLONES REACTING AGAINST AUTOLOGOUS HUMAN MELANOMA

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Peripheral blood lymphocytes (PBL) of a melanoma patient were cultivated with autologous melanoma cells (Auto-Me) and recombinant interleukin 2 (RIL-2, Biogen)

for 30 days. In these conditions, PBL proliferated but did not develop cytotoxic activity against Auto-Me and K562 cells. The phenotype of PBL at the 30th day was 95% CD3, 95% CD4, 1% CD8 and 0% CD16. Clones were then derived at 1 cell/well in the presence of irradiated Auto-Me, RIL-2 (25 U/ml) and Daudi cells as feeder. The 81 growing clones were screened for cytotoxicity and proliferating activity in the presence of Auto-Me. Twelve clones were cytotoxic for Auto-Me and 22 clones showed significant proliferation with Auto-Me; 67 clones exhibited both cytotoxic and proliferating activity. Preliminary results of the specificity analysis showed that one clone which proliferated to Auto-Me expressed cytotoxicity on Auto-Me but not on 12 different targets including autologous EBV-B cells and fibroblasts, 6 allogeneic melanomas, 2 lymphoblastoid lines, Daudi and K562.

Supported by Grant N.86.00663.44 of Consiglio Nazionale delle Ricerche, Rome, Italy.

BIOCHEMICAL EVALUATION OF HYPERCALCAEMIA ASSOCIATED WITH BRONCHOGENIC CANCER

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Bronchogenic carcinoma (BC) may be frequently associated with hypercalcaemia, due to direct bone resorption by tumour cells, or alternatively by osteoclastic bone resorption stimulated by humoral factors (i.e. parathyroid hormone (PTH)-like substances and prostaglandins (PGE)).

In 160 patients with BC, hypercalcaemia was found in 19 cases (11.8%). By measuring the biological parameters of PTH activity and the circulating levels of PTH and PGE the hypercalcaemic patients were divided into three groups. The first group (n=6) presented the typical biochemical pattern of hyperparathyroidism, the second group (n=10) was characterized by high circulating levels of PGE; in the third group (n=3) all the parameters considered were normal. A metastatic bone involvement as evidenced by radiologic and scintigraphic means, was documented only in patients of the first and second group. These data further emphasize the importance of humoral factors in the pathogenesis of hypercalcaemia associated with BC.

NUCLEIC ACID BINDING OF TRANS-4-AMINOSTILBENE DERIVATIVES *IN VITRO* AND *IN VIVO*

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Trans-4-acetylaminostilbene (AAS) is a strong tumour initiator in rat liver. The model ultimate carcinogen, N-acetoxy-AAS, reacts with nucleic acids *in vitro* and predominantly with guanine. The major adducts are four cyclic isomers in which guanine is substituted at N2 and N3 as shown by reactions with Guo and d-Guo. In addition, two minor guanine adduct fractions were identified and shown to consist also of sets of isomers. After oral administration of trans-4-dimethylaminostilbene (DAS) and AAS, the cyclic adducts and presumably also the minor adducts are formed in rat liver DNA. Substitution of guanine at N2 and N3 labilizes the glycosidic bond, which results in depurinization of DNA, and may impair nuclease activity, which could explain the observed incomplete hydrolysis of modified DNA. Experiments with AAS labelled in the acetyl group indicate that non-acetylated adducts are also generated in liver RNA and DNA to some extent. Among these non-acetylated adducts the cyclic guanine adducts are also present. A number of persistent adducts could be demonstrated 28 days after oral administration of DAS.

CORRELATION OF ELASTOSIS WITH SOME MORPHOPATHOLOGICAL PROGNOSTIC FACTORS IN BREAST CARCINOMAS

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The increase of elastic tissue was found on haematoxylin-eosin stained tumour sections in 54 (22%) from 245 invasive ductal carcinomas of the breast treated by radical mastectomy, as a pink, homogeneous or finely fibrillar sheaths around carcinomatous ducts and focal deposits in contact with tumour parenchyma. The elastosis was assessed subjectively in three degrees and analysed in relation to the tumour size, histological pattern, grade of malignancy, lymph node metastases and age of patients. The percentage of elastin positive tumours increased in parallel with their histological differentiation from trabecular carcinomas to pure glandular carcinomas. Elastosis correlated to some extent with the grade of malignancy - a higher proportion of elastin positive tumours was found in low grade group and conversely, a higher proportion of cases without elastosis in high grade tumours. No relation was established between elastosis