tumours failed to respond because prolactin was not abolished; (5) 2/2 oestrogen dependent tumours got worse on oestrogens; (6) 2/2 androgen dependent tumours got worse on testosterone; (7) 10/11 *in vitro* independent tumours failed to respond. The *in vitro* test thus gave a correct prediction in 37 of 40 patients.

CHANGES IN RESPONSE TO CHEMOTHERAPEUTIC AGENTS DURING THE LIFE HISTORY OF MONOLAYER CULTURES OF A MOUSE TUMOUR CELL LINE. P. R. TWENTYMAN and N. M. BLEEHEN. Academic Department of Radiotherapy, Middlesex Hospital Medical School, London.

At the previous meeting of the Association, we described how EMT6 mouse tumour cells become less sensitive to bleomycin as they pass from exponential growth into plateau phase. This result was the opposite of that reported by other workers using Chinese hamster cells. Detailed investigation of the proliferation kinetics of our EMT6 cell line has revealed that the plateau phase may be subdivided into early plateau (with a pulse labelling index of 25% and considerable cell loss) and late plateau (with a labelling index of $<5\%$ and little cell loss). Sensitivity to bleomycin is indeed reduced during early plateau (compared with exponentially growing cells), but in late plateau the sensitivity becomes greater than that in exponential cells. Sensitivity to a number of other chemotherapeutic agents has also been investigated in cultures of various ages.

THE EFFECT OF WHOLE BODY HYPERTHERMIA IN ADVANCED CANCER. R. T. PETTIGREW, C. M. LUDGATE and A. N. SMITH. Department of Anaesthetics and Department of Clinical Surgery, Western General Hospital and University of Edinburgh.

The anaesthetized patient is immersed in molten wax at 50°C. This reverses the normal physiological processes of heat loss. A 5°C rise is achieved in one hour and maintained for 3–4 h. Fifty-five patients with advanced cancer have been heated to 41·8°C and the tumour response assessed by criteria which include relief of pain, weight gain, serial biopsy changes and evidence of tumour regression. One group (45 patients) was treated by hyperthermia alone; sarcomata and gastrointestinal tract tumours were the most responsive (8 in 11); an intermediate group, skin and lung tumours, was less so (6 in 16); a third (mainly genito-urinary), the least (0 in 14). The addition of chemotherapy to hyperthermia in 10 patients raised the proportion regressing from 11 in 23 (48%) to 7 in 10 (70%). Occasional complications were mild superficial burns, tracheitis, ventricular fibrillation and disseminated intravascular coagulation.

ANALYSIS OF THE ANTIMETASTATIC ACTION OF THE ANTIMITOTIC AGENT ICRF 159. K. HELLMANN, S. E. JAMES and A. J. SALSBURY. Imperial Cancer Research Fund, London.

ICRF 159 inhibits metastases from the spontaneously metastasizing Lewis lung carcinoma (3LL) without markedly impeding the growth of the primary implant. It has previously been proposed that this antimetastic action of ICRF 159 is due to the inhibition of malignant cell release from the primary tumour consequent upon normalization of the tumour blood vessels by the drug. Lung "metastases" due to intravenous injection of 3LL cells should therefore be unaffected by ICRF 159 administration. This was not, however, found to be the case.

When primary tumours were excised up to 6 days following implantation, secondary growths were not apparent in the lungs at 21 days. Treatment of primary tumours for the first 6 days by 30 mg/kg ICRF 159, at a time therefore when no circulating malignant cells would have been present, produced an almost complete inhibition of metastases. Thus, under these conditions the antimetastatic action cannot be ascribed to an effect of the drug on 3LL cells in the blood stream.

A COMPARISON OF THE CELL KILLING IN THE MOUSE AFTER EXPOSURE TO FTORAFUR AND TO 5-FLUOROURACIL. L. M. VAN PUTTEN, L. K. J. KRAM-IDSENGA and M. PIJPERS-DE BRUIN. Radiobiological Institute TNO, Rijswijk, Holland.

Ftorafur (N-1-(furanidyl)-5 fluorouracil) was compared with 5-fluorouracil (5-FU) in mice. The LD_{50} and the slopes of the dose-effect curves for killing of L1210 leukaemia

cells and of haemopoietic stem cells in the bone marrow indicate that Ftorafur is 2–3 times less effective than 5-FU. However, it is about 7 times less effective in killing rapidly proliferating haemopoietic stem cells. This finding is hard to explain since Ftorafur is reported to be active only through release of 5-FU, a report which is in agreement with our observation of ineffectiveness of Ftorafur upon incubation *in vitro* in concentrations 10 times higher than effective 5-FU concentrations. On the other hand, the lower toxicity of Ftorafur for proliferating stem cells might explain the reported better tolerance upon prolonged administration to patients in comparison with equally effective doses of 5-FU. A study, made to verify this, has indicated that upon fractionated administration Ftorafur is not only much less toxic than 5-FU but also less effective in killing L 1210 cells.

If conditions in man are comparable with those in mice, part of the good haematological tolerance to prolonged administration of Ftorafur may be due to a decrease in the fraction of the drug being hydrolyzed to liberate 5-FU.

RECENT RESULTS WITH CARCINOGEN BINDING PROTEINS. B. KETTERER and E. TIPPING. Middlesex Hospital Medical School. D. BEALE. ARC Institute of Animal Physiology. B. ABRAHAM and J. MEUWISSEN. Katholieke Universiteit te Leuven, Belgium.

Three azodye carcinogen binding proteins have been purified in our laboratories with sedimentation coefficients of 4·7, 3·5 and 1·7S. The 4·7S protein has a pI of 9·0, a low tyrosine and a high CyS content, a complex absorption spectrum for bound azodye and readily dissociates into subunits.

The 3·5S protein, ligandin, is a relatively small molecule composed of two apparently identical subunits of MW 23,000. It nevertheless binds ligands as diverse as oestrone sulphate, bilirubin and haematin with affinity constants for the first binding site of 10^6 to at least 10^8.

The abundance and wide tissue distribution of ligandin make it of particular interest. An apparent connection with drug metabolizing enzymes indicates that it is more than an intracellular equivalent of serum albumin.

LACK OF SYNERGY BETWEEN N-METHYL-N-NITROSOUREA (MNU) AND CYCLOPHOSPHAMIDE (CP) IN RAT URINARY BLADDER. J. ST. J. WAKEFIELD and R. M. HICKS. Middlesex Hospital Medical School, London.

A single, intravesicular dose of MNU is non-carcinogenic in the normal life span of the rat. By contrast, 4 bi-weekly doses produce bladder tumours from 15 weeks on (Hicks and Wakefield, *Chem-Biol. Interact.*, 1972, **5**, 139). Each dose is followed by necrosis then hyperplasia of the epithelium. Intraperitoneal injection of CP also causes necrosis followed by hyperplasia of the bladder epithelium, but no tumours develop after multiple (12) doses. This suggests that prolonged hyperplasia *per se* is not carcinogenic in the absence of some further stimulus.

Rats given either a single dose of MNU followed by CP, or CP followed by a single dose of MNU, also failed to develop tumours. These results show no co-carcinogenesis with MNU and CP, even though the target tissue for both compounds is the same.

SYNCARCINOGENESIS WITH N-METHYL N-NITROSOUREA (MNU) AND CYCLAMATE IN RAT URINARY BLADDER. J. CHOWANIEC, J. ST. J. WAKEFIELD and R. M. HICKS. Middlesex Hospital Medical School, London.

Cyclamate is suspect as a bladder carcinogen, but reports from different experimentalists are conflicting. In this laboratory, only one animal on a cyclamate containing diet has so far developed a bladder tumour in the absence of any other treatment.

One intravesicular dose of MNU is not carcinogenic but 4 doses are (Hicks and Wakefield, *Chem-Biol. Interact.*, 1972, **5**, 139). Animals which have received one intravesicular dose of MNU are now being maintained on a cyclamate containing diet. Of these, 21 animals have been killed so far and 9 had bladder tumours. These results demonstrate syncarcinogenesis with MNU and cyclamate in the bladder. By contrast, co-carcinogenesis could not be demonstrated with MNU and the cytotoxic, but not carcinogenic, cyclophosphamide (see previous abstract, J. St. J. Wakefield and R. M. Hicks). We suggest that cyclamate may be a weak bladder carcinogen, not normally effective in the life span of the animal.