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ENHANCEMENT OF ANTI-CANCER ACTIVITY OF CYTOTOXIC CHEMOTHERAPY WITH PROTECTION OF NORMAL TISSUES BY INHIBITION OF P.G.* SYNTHESIS

T.J. Powles, P. Alexander, J.L. Millar

Institute of Cancer Research and
Royal Marsden Hospital, Sutton, Surrey.

In experimental systems we find that inhibition of P.G. synthesis by I. and F.P. can affect the anti-proliferative activity of alkylating agents differently in tumours and normal tissues. In one tumour system studied, F.P. and I. rendered a resistant tumour more responsive to cytotoxic therapy. The table summarizes one of three experiments in which inhibition of growth of a chemoresistant variant of the Walker tumour by chlorambucil, at a level well below the toxic dose, was achieved by the concurrent administration of I. or F.P. While the standard Walker tumour

TABLE Influence of P.G. synthesis inhibitors on the anti-tumour activity of chlorambucil on a chemo-resistance line of Walker tumour*

Treatment@	Total No.	Day of death							
None	4	9	9	9	9				
CHL	8	9	9	9	9	10	10	10	11
I	8	9	9	9	9	9	9	9	10
FP	8	9	9	9	9	9	10	12	13
CHL + I	8	9	10	10	13	14	15	>30	>30
CHL + FP	8	10	12	13	13	>30	>30	>30	>30

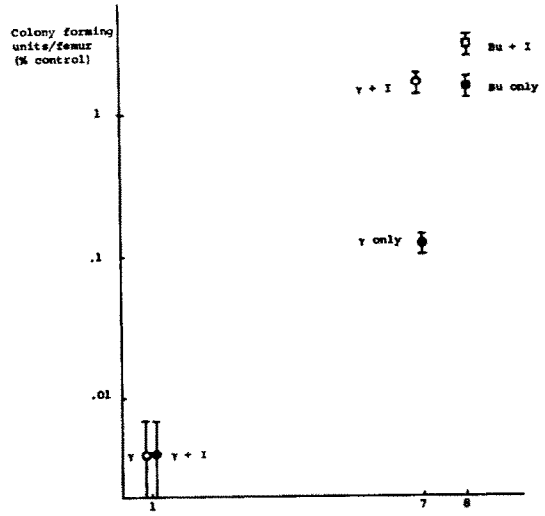
* 10^7 ascites tumour cells inoculated i.p. into 12 week old Wistar rats.

@ Tumour inj. Day 0
CHL chlorambucil 10 mg/kg single i.p. inj. 24 hr. after tumour.
I Indomethacin inj. s.c. 7 mg/kg three times per day Day 0, 1 and 2.
FP Flurbiprofen inj. s.c. 7 mg/kg three times per day Day 0, 1 and 2.

* Abbreviations: P.G. prostaglandin; I., indomethacin; F.P. flurbiprofen (kindly provided by the Boots Co. Ltd.)

is curable by chlorambucil (1) the resistant line used in these experiments was induced by repeated transplantation in the presence of low doses of chlorambucil.

In the treatment of cancer the dose-limiting toxicity of alkylating agents and of whole-body radiation is largely the destruction of stem cells in the bone-marrow and the gut and to be of practical value potentiation must be selective. The data which has so far been obtained shows that the P.G. inhibitor, I. does not cause increased destruction of stem cells by these agents but may even promote their recovery. The figure illustrates this effect following whole-body irradiation with 900r of γ-rays. The number of stem cells in the bone-marrow was assessed, as previously described (2), at different times after irradiation by injecting i.v. the marrow from one femur or one-fifth of a femur into a heavily irradiated recipient and then 7 days later counting the number of colonies of haemopoetic cells in the spleen. The initial destruction by γ-rays of the stem cell pool was the same in control as in I-treated mice but 7 days after



Days after cytotoxic treatment.

● 900r γ-rays to CBA mice; marrow from one femur removed immediately or 7 days after irradiation and injected i.v. into heavily irradiated mice (Colony forming units (CFU) measured by spleen colony assay (2)).

○ As above plus 1.75 mg/kg of I s.c. three times per day for 3 days starting one day before whole-body irradiation.

■ 40 mg/kg busulphan i.p. at time 0: CFU measured 8 days later.

□ 40 mg/kg busulphan plus 1.75 mg/kg I s.c. three times per day for 3 days starting one day before Bu.

irradiation the number in the mice that had received I. was more than 10 times greater. This suggests that inhibition of P.G. synthesis hastens the recovery of bone-marrow stem cells in the irradiated host. A similar, though smaller, effect was also seen when bone-marrow aplasia was induced by the alkylating agent busulphan (see Figure).

Though it is possible to enumerate the colony-forming cells in the gut (3), the effect of I. on gut damage by alkylating agents has so far only been determined in a qualitative way by assessing its effect on the mortality induced in CBA mice that had received 20 mg/kg of the alkylating agent melphalan as a single i.p. injection. Millar et al. (4) found that the alkylating agent melphalan at this dose kills CBA mice within 5 days as a result of necrosis of the mucosa of the small gut. In three experiments 1.75 mg/kg of I. given three times per day for three days starting on the day before the melphalan was given had a protective effect. In the control groups $^{8}/_{10}$, $^{10}/_{10}$ and $^{10}/_{10}$ of the mice died, whereas in the I-treated group the mortality was $^{4}/_{10}$, $^{5}/_{10}$ and $^{5}/_{10}$. As in the case of the bone-marrow I. reduced damage to the gut though whether this occurs by reducing the initial kill or by promoting recovery remains to be established.

The mechanisms by which inhibitors of P.G. synthesis enhance the destruction of at least one type of tumour by alkylating agents while promoting the recovery of normal tissues remains to be elucidated. We are exploring the possibility that high levels of endogenous P.G. synthesis by some tumours may be involved in chemoresistance.

The divergent effects of I. and F.P. on the response of normal and tumour cells to alkylating agents suggests obvious therapeutic application and currently the effect of adding F.P. to chemotherapy is being investigated in patients with metastatic breast cancer who have failed to respond to chemotherapy only. So far, 3 such patients have been treated in this way and in every one an improved response was observed when the chemotherapy was administered with F.P.

REFERENCES

1. J.L. Everett, J.J. Roberts and W.C.J. Ross, J. Chem. Soc. 2386 (1953).

2. J.L. Millar, N.M. Blackett and B.N. Hudspith, Cell and Tissue Kinetics, in the press.

3. H.R. Withers and M.M. Elkind, Int. J. Radiat. Biol. 17, 261 (1970).

4. J.L. Millar, B.N. Hudspith, T.J. McElwain and T.A. Phelps (1978) (submitted for publication).