J. Pharm. Pharmac., 1972, **24**, 575

LETTERS TO THE EDITOR

Non-steroidal anti-inflammatory drugs and leucocyte emigration

It has been recently suggested that the success of carrageenan foot oedema for the assay of antirheumatic drugs is mainly due to the marked cellular migration which occurs during this type of acute inflammation (Di Rosa & Willoughby 1971a; Di Rosa, 1972). On this model non-steroidal anti-inflammatory drugs suppress mainly the last phase of the response, namely the "prostaglandin phase", in addition their ability to suppress this phase correlates directly with their ability to prevent mononuclear leucocyte migration into the inflamed area (Di Rosa & Willoughby 1971b; Di Rosa, Papadimitriou & Willoughby, 1972). Thus the measure of oedema inhibition is also a rough measure of inhibition of cell migration.

We have attempted to expand these observations and to obtain more reproducible and quantitative data on the effects of non-steroidal anti-inflammatory drugs on leucocyte migration by using dextran pleurisy in the rat as the model of acute inflammation.

Dextran has been preferred to other irritants because it induces about the same magnitude of migration of both polymorphs and mononuclears, although at different times (Hurley, Ryan & Friedman, 1966).

Male albino rats of the Wistar strain, 120–130 g, were injected with 1 ml of saline containing 6% dextran (mol. wt., 80 000) into the pleural space. At 4, 14 or 24 h after injection, intrapleural exudate was collected and total and differential leucocyte counts were made according to Hurley & others (1966). Four representative non-steroidal anti-inflammatory drugs were tested: indomethacin, phenylbutazone, aspirin and flufenamic acid. Each drug was administered by mouth 1 h before dextran injection.

Fig. 1 shows that intrapleural injection of dextran caused the leucocytes to migrate into the pleural space. Magnitude and time of the peak responses exhibited by polymorphs (about 20×10^6 cells at 4 h) and by mononuclears (about 15×10^6 cells at 24 h) were in the same range of the respective responses as that reported by Hurley & others (1966). Total leucocyte migration at 4 h was virtually unaffected by a treatment with indomethacin (10 mg/kg) or phenylbutazone (90 mg/kg) while both drugs caused a notable reduction of the cell migration at 14 and 24 h. This effect was clearly due to the marked suppression of mononuclear migration which at 14 and 24 h was reduced by more than 50% of that of control rats. In contrast, polymorph migration was only slightly affected, the observed inhibition being about 10–15% at 4 h, and of equivalent degree subsequently.

Similar results were observed after aspirin (270 mg/kg) or flufenamic acid (270 mg/kg).

Inhibition of mononuclear migration into the inflamed area, resulting from treating rats with antirheumatic drugs, was dose dependent. Calculated ED50 values (mg/kg) were: indomethacin 7·5; phenylbutazone 60; aspirin and flufenamic acid 180.

The potency ratio exhibited by indomethacin, phenylbutazone and aspirin in suppressing mononuclear migration closely agrees with the effectiveness of the drugs on various models of inflammation as well as with their clinically established anti-rheumatic activity. Flufenamic acid however appeared to be less potent in its effec-

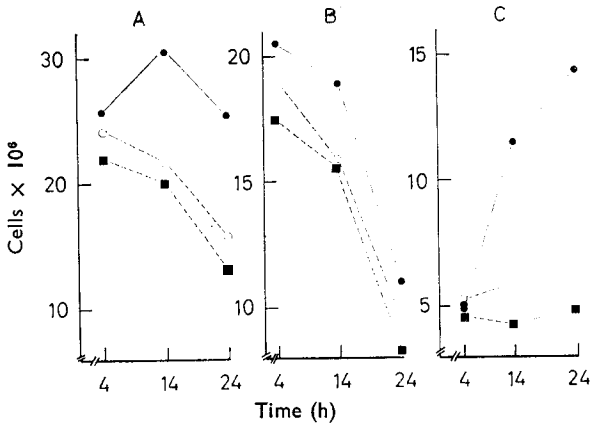

FIG. 1. Total leucocytes (A), polymorphs (B) and mononuclears (C) counts in rat pleural exudates at various times after intrapleural injection of 1 ml of 6% dextran in saline. Each point represents the mean value of 6–8 rats. Controls ●, indomethacin ○, phenylbutazone ■. Pleural cavity from normal animals, i.e. not injected with dextran, contained on the average about 5×10^6 cells, virtually all mononuclear ($\geq 95\%$).

tiveness in laboratory tests, in which it lies between indomethacin and phenylbutazone (Winter, 1965) than in its antirheumatic activity, which lies between phenylbutazone and aspirin (Camus, 1968).

The poor susceptibility of polymorphs to antirheumatic drugs could explain the failure of indomethacin in preventing leucocyte emigration into dogs knee injected with sodium urate, this irritant in fact causes cellular migration mainly sustained by polymorphs (Van Arman, Carlson & others, 1970).

Our results demonstrate a different susceptibility to non-steroidal anti-inflammatory agents of the two main type of cells involved in acute inflammation, namely polymorphs and mononuclears.

It is also significant that antirheumatic drugs are almost specifically effective in preventing migration of mononuclears, the cells responsible for chronic processes.

It has been shown recently that non-steroidal anti-inflammatory drugs inhibit prostaglandin synthesis and release (Vane, 1971; Ferreira, Moncada & Vane, 1971; Smith & Willis, 1971 as well as some effects of prostaglandins *in vitro* (Sorrentino, Capasso & Di Rosa, 1972). Our data do not preclude the interference by antirheumatic drugs in the actions of prostaglandins, rather, they suggest that the roles played by prostaglandins and mononuclear cells in inflammation are closely related.

Institute of Pharmacology,
School of Medicine,
University of Naples, Italy.

M. DI ROSA
L. SORRENTINO
L. PARENTE

March 24, 1972

REFERENCES

CAMUS, J. P. (1968). *Press Med.*, **76**, 1071–1072.
DI ROSA, M. (1972). *J. Pharm. Pharmac.*, **24**, 89–102.
DI ROSA, M., PAPADIMITRIOU, J. & WILLOUGHBY, D. A. (1971). *J. Path.*, **105**, 239–256.
DI ROSA, M. & WILLOUGHBY, D. A. (1971a). *J. Pharm. Pharmac.*, **23**, 297–298.
DI ROSA, M. & WILLOUGHBY, D. A. (1971b). *Naunyn-Schmiedebergs Arch. Pharmak.*, **269**, 482.
FERREIRA, S. H., MONCADA, S. & VANE, J. R. (1971). *Nature New Biology*, **231**, 237–239.
HURLEY, J. V., RYAN, G. B. & FRIEDMAN, A. (1966). *J. Path.*, **91**, 575–587.
SMITH, J. B. & WILLIS, A. L. (1971). *Nature New Biology*, **231**, 235–237.
SORRENTINO, L., CAPASSO, F. & DI ROSA, M. (1972). *Eur. J. Pharmac.*, **17**, 306–308.

VAN ARMAN, C. G., CARLSON, R. P., RISLEY, E. A., THOMES, R. H. & NUSS, G. W. (1970). *J. Pharmac. exp. Ther.*, **175**, 459–468.

VANE, J. R. (1971). *Nature New Biology*, **231**, 232–235.

WINTER, C. A. (1965). *Non-steroidal anti-inflammatory drugs*, p. 190–202. Editors: Garattini, S. & Dukes, M. N. G. Amsterdam, Excerpta Medica.

The effect of chlorpromazine on drug metabolism

Curry, Lader & others (1971) have demonstrated a lowering of the steady-state plasma level of chlorpromazine, beginning two weeks after the start of treatment with a constant dose of drug. They suggest that this might result from induction of liver drug-metabolizing enzymes. Certainly, it has been shown that chlorpromazine causes enzyme induction in animals (Conney, 1967). However, in a recent report by Gram & Overø (1972) there are indications that chlorpromazine inhibits the metabolism of imipramine in man. In our work, we have examined the effect of chlorpromazine on drug-metabolizing capacity in two situations—firstly, in schizophrenic patients receiving this drug and secondly, using isolated liver preparations from rats treated with chlorpromazine and, for comparison, with barbitone.

Drug-metabolizing capacity in chlorpromazine-treated patients, in young control subjects and in elderly drug-free patients was assessed using the plasma antipyrine half-life technique, the dose, time of sampling and antipyrine estimations being as previously described (O'Malley, Crooks & others, 1971). Patients included in the study had been receiving chlorpromazine (150–600 mg) for at least 2 months and had not been given other drugs in that time. The phenothiazine was stopped approximately 10 h before ingestion of the test drug. Details of the animal study are as outlined in Table 2.

From Table 1 it can be seen that the mean plasma antipyrine half-life in the chlorpromazine-treated patients was longer than in the younger controls ($P < 0.001$; students t-test) and was the same as that found in the elderly control subjects. Had enzyme induction occurred in the drug-exposed group a lower plasma antipyrine

Table 1. *Plasma antipyrine half-life values in three groups of subjects.*

Subjects	No.	Age	Plasma antipyrine half-life (h)
Young	61	26·0 ± 2·5	12·0 ± 3·5
Elderly	18	77·6 ± 8·7	17·4 ± 6·8
Chlorpromazine treated ..	10	61·5 ± 9·3	17·7 ± 4·9

Results are shown as means ± s.d.

Table 2. *Hexobarbitone oxidation by isolated liver preparations from control and treated animals.*

	μmol hexobarbitone oxidized/g wet wt liver per h
Control	0·84 ± 0·11
Barbitone (10 mg/kg daily)	0·95 ± 0·14
Chlorpromazine (10 mg/kg daily)	0·89 ± 0·17
Barbitone (50 mg/kg daily)	2·65 ± 1·10
Chlorpromazine (50 mg/kg daily)	1·43 ± 0·42

Hexobarbitone oxidation was measured using [^{3}H]-labelled hexobarbitone as substrate. Animals were treated for four days as indicated, the drugs being given in two daily doses. Hexobarbitone oxidation was measured on the fifth day using liver 9000 *g* supernatants. Results are expressed as means and s.d. for groups of six animals.