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Studies on the anti-inflammatory properties of dapsone in a variety of animal models

D.K. GEMMELL, A.J. LEWIS & W.H. STIMSON

Scientific Development Group, Organon Laboratories Limited, Newhouse, Lanarkshire and Department of Biochemistry, University of Strathclyde, Glasgow, Scotland

Dapsone (4,4'-diaminodiphenylsulphone) is an established antimalarial and antileprotic agent (Goodman & Gilman, 1975) that is also currently used for the treatment of certain bullous diseases (Alexander, 1975). The demonstration that it inhibited both lysosomal enzyme activity (Barranco, 1974; Mier & van den Hurk, 1975) and complement activation (Milliken & Conway, 1974) suggested that it might be of value in the treatment of inflammatory diseases. Indeed, Williams, Capstick, Lewis & Best (1976) have recently reported that dapsone possesses anti-inflammatory activity in the carrageenin paw oedema, and adjuvant arthritis, and that furthermore a trial in man has demonstrated that dapsone successfully treated rheumatoid arthritis (McConkey, Davies, Crockson, Butler & Constable, 1976).

We have now examined the anti-inflammatory activity of dapsone in additional models of inflammation. Male Wistar rats (CE/CFHB, Carworth Europe) or female Dunkin Hartley guinea pigs, have been used throughout the following experiments and the drug vehicle used was 5% mulgofen. A single oral dose of dapsone (ED_{50} 54 mg/kg) was as effective as phenylbutazone (ED_{50} 58 mg/kg) in inhibiting the 4 h oedema produced in the rat paw by kaolin administration (0.1 ml, 10% suspension). The anti-inflammatory activity of dapsone is not mediated via the pituitary-adrenal axis since adrenalectomy did not affect its oral activity in the kaolin paw oedema. In addition, dapsone possesses local anti-inflammatory activity measured 4 and 24 h after administration to rats via the subplantar route in combination with a 5% kaolin suspension (min. effective dose 3 mg/paw).

In the guinea pig model of u.v. induced erythema (Winder, Wax, Burr, Been & Rosiere, 1958) oral administration of dapsone 1 h prior to irradiation was approximately 10 times less effective than phenylbutazone (ID_{50} 16 mg/kg) in delaying the erythema measured 4 h post irradiation. However, in a model of reversed passive Arthus in the rat (after Cochrane, 1965), produced by i.v. challenge of egg albumin after prior sensitization into the footpad with rabbit anti-egg albumin, dapsone was approximately 6 times more effective than phenylbutazone administered orally 1 h before challenge.

Since acute gastric irritation is a common feature of most non-steroidal anti-inflammatories gastric damage was assessed in starved rats. Dapsone produced no clear gastric (or intestinal) irritation 6 h after oral administration at doses as high as 600 mg/kg. This contrasts with the marked irritant effects of phenylbutazone in this test at doses as low as 50 mg/kg.

After both prophylactic (0–14 d) and therapeutic (14–21 d) oral administration 150 but not 100 mg kg^{-1} d^{-1} dapsone significantly suppressed the adjuvant induced secondary inflammation (0.6 mg M. butyricum in 0.05 ml paraffin oil) on day 14 and day 21, respectively. The primary inflammation was not affected at either time interval. The minimum effective dose of orally administered phenylbutazone against the secondary lesions was 30 mg/kg per day. The cyanotic appearance of these dapsone-treated rats may correspond to the cyanosis attributed to methemoglobinemia reported during dapsone therapy in man (Goodman & Gilman, 1975). No gastric lesions were observed in any of these animals treated daily with dapsone.

Dapsone ($>10^{-5}$ M) inhibited zymosan-induced release of lysosomal enzyme (β-glucuronidase) from cultured rat peritoneal macrophages (Stimson, Hunter & Manos, 1977). This was not due to a reduction in phagocytosis of the zymosan but a part of the apparent membrane stabilizing activity was caused by direct inhibition of enzyme activity.

In conclusion, these results partly confirm and extend the observations that dapsone exhibits anti-inflammatory activity in animal models of inflammatory processes. Whether dapsone will prove an effective therapy in inflammatory diseases such as rheumatoid arthritis awaits further clinical evidence. In this respect the common side effects of dapsone therapy, such as methemoglobinemia and haemolysis, will have to be carefully considered against its clinical benefits.

References

ALEXANDER, J.D. (1975). *Dermatitis Herpetiformis.* pp. 285–312. London, Philadelphia and Toronto: W.B. Saunders Co. Ltd.

BARRANCO, V.P. (1974). Inhibition of lysosomal enzymes by dapsone. *Arch. Derm.*, **110**, 563–566.

COCHRANE, C.G. (1965). The Arthus reaction. In *Inflammatory Process.* ed. Zweifach, B., Grant, L. & McLusky, R. pp. 613–648. New York: Academic Press.

GOODMAN, L.S. & GILMAN, A. (1975). *The Pharmacological Basis of Therapeutics*. pp.1216–1218. London: Heinemann.

McCONKEY, B., DAVIES, P., CROCKSON, R.A., BUTLER, M. & CONSTABLE, T.J. (1976). Dapsone in rheumatoid arthritis. *Rheum. Rehabil.*, **15**, 230–234.

MIER, P.D. & VAN DEN HURK, J.J.M.A. (1975). Inhibition of lysosomal enzymes by dapsone. *Br. J. Derm.*, **93**, 471–472.

MILLIKEN, L.E. & CONWAY, F.R. (1974). Effect of drugs on the pillemer pathway–dapsone. *J. invest. Derm.*, **62**, 541.

STIMSON, W.H., HUNTER, I.C. & MANOS, C. (1977). Comparison of the effects of human pregnancy serum and anti-inflammatory compounds on the release of lysosomal enzymes from macrophages. *Br. J. exp. Path.* (in press).

WILLIAMS, K., CAPSTICK, R.B., LEWIS, D.A. & BEST, R. (1976). Anti-inflammatory actions of dapsone and its related biochemistry. *J. Pharm. Pharmac.*, **28**, 555–558.

WINDER, C.V., WAX, J., BURR, V., BEEN, M. & ROSIERE, C.E. (1958). A study of pharmacological influences on ultraviolet erythema in guinea pigs. *Arch. int. Pharmacodyn.*, **116**, 261–292.

Effects of atropine and pimozide on hypothermia induced by apomorphine or oxotremorine in rats

B. COX & T.F. LEE

Department of Pharmacology, Materia Medica and Therapeutics, Manchester University

Injection of apomorphine (1.25 to 20 μg) or dopamine (5 to 20 μg) into the preoptic-anterior hypothalamus of the rat caused a dose related fall in body temperature, which was reversed by pretreatment with pimozide (0.5 mg/kg i.p.; Cox & Lee, 1977)

In this study we have tested the specificity of the blockade by pimozide by comparing its effects on apomorphine-induced hypothermia with those on the hypothermia induced by the muscarinic agonist oxotremorine. The ability of atropine to antagonize apomorphine or oxotremorine-induced hypothermia has also been tested.

Two types of experiment were performed. In the first series the effects of unilateral intrahypothalamic injection of apomorphine and oxotremorine were compared in control rats and in rats pretreated systemically with either atropine or pimozide. In the second series the agonists were injected by the systemic route and the antagonists were given by bilateral intrahypothalamic injection. Intrahypothalamic drug injections were made in a dose volume of 1 μl through previously implanted guide cannulae. Core temperature was measured with a rectal thermistor probe at an ambient temperature of 17 ± 1°C.

The hypothermia after intrahypothalamic injection of apomorphine was reversed by systemic pimozide ($P < 0.01$) but unaffected by systemic atropine (Table 1). Conversely intrahypothalamic oxotremorine was significantly antagonized by atropine, but not by pimozide. Similar results were obtained when the routes of injection of agonist and antagonist were reversed.

Table 1 Effect of atropine and pimozide on hypothermia induced by apomorphine or oxotremorine in rats

	Mean change in core temperature (°C ± s.e. mean)			
Drug	*Saline i.p.*	*Pimozide (0.5 mg/kg i.p.)*	*Atropine (0.5 mg/kg i.p.)*	*Atropine (2.5 mg/kg i.p.)*
Apomorphine (10 μg i.h.)	−0.9 ± 0.2	+0.4 ± 0.1**	−0.7 ± 0.2	−0.9 ± 0.2
Oxotremorine (1.25 μg i.h.)	−0.5 ± 0.05	−0.3 ± 0.2	−0.2 ± 0.03*	−0.08 ± 0.06**
	Saline i.h.	*Pimozide (0.5 μg i.h.)*[1]	*Atropine (0.5 μg i.h.)*[1]	*Atropine (2.5 μg i.h.)*[1]
Apomorphine (1.25 mg/kg i.p.)	−1.8 ± 0.2	−0.9 ± 0.2*	−1.4 ± 0.3	−1.5 ± 0.27
Oxotremorine (0.25 mg/kg i.p.)	−3.1 ± 0.3	−2.5 ± 0.1	−1.8 ± 0.3*	−1.3 ± 0.11**

[1] Injection made bilaterally into the hypothalamus (i.h.), n = between 3 and 13 observations, *$P < 0.05$, ** $P < 0.01$ Mann-Whitney U test.