Olac rats (approximately 200 g weight) were lightly anaesthetized with ether and injected intravenously with ^{125}I albumin (5 μCi) and Evans blue (40 mg/kg). Drugs were then injected in 0.1 ml saline intradermally into the shaved abdominal skin. Twenty minutes later, the animals were killed and the skin removed. Discs of skin were excised which included the whole of the lesion (as indicated by blueing). The excised skin was wrapped in a single layer of parafilm and the radioactivity measured in a gamma counter. The radioactivity in 10 μl of blood was measured as a reference.

Intradermal injection of PG E_1, E_2 and D_2 produced increases in vascular permeability. These effects were reduced by admixture with PGD_1 (Table 1).

PGD_1 did not modify the increased vascular permeability induced by histamine or bradykinin, as would be expected if it was acting through vasoconstriction. $PGF_{2\alpha}$ reduced the effects of histamine and bradykinin as well as prostaglandins E_1, E_2 and D_2 showing that its effect was less specific than that of PGD_1. These results suggest a role for PGD_1 and $PGF_{2\alpha}$ as modulators of skin inflammation.

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A comparison of the histopathological effects of anaphylatoxin (AT) and prostaglandins E_2 (PGE_2) and $F_{2\alpha}$ ($PGF_{2\alpha}$) in guinea-pig lungs

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In a previous study it was shown that AT induced the release of a substance with prostaglandin-like activity in isolated perfused guinea-pig lungs (Sackeyfio, 1972). Other investigations have also shown that the complex effects of AT in the guinea-pig lead not only to the release of the bronchoconstrictor substance, histamine (Sackeyfio, 1971) but also to the stimulation of adrenergic mechanisms which are predominantly bronchodilator (Hicks & Sackeyfio, 1971). However, the dominant symptomatic feature of (AT) administration in the guinea-pig is intense respiratory distress. The present investigation was undertaken with the view that a comparison of the histopathological effects of AT with those of known or suspected mediators of AT activity might provide further elucidation of the mechanisms involved in AT activity.

AT was prepared as previously reported (Sackeyfio, 1971). Groups of guinea-pigs were anaesthetized with pentobarbitone (60 mg/kg i.p.) and injected i.v. with one or other of the following: AT (0.5 ml/kg); rat native serum (0.5 ml/kg), histamine (20 μg/kg), PGE_2 (1 mg/kg) or $PGF_{2\alpha}$ (1 mg/kg). The lungs were isolated, fixed in 10% buffered formalin for 24 h, embedded in wax, sections were stained with haematoxylin and eosin and examined microscopically. Lungs from untreated guinea-pigs were similarly prepared for comparison.

Examination of the slides showed that AT, histamine, PGE_2 and $PGF_{2\alpha}$ induced varying degrees of bronchoconstriction and thickening of the bronchial muscle (the order of intensity being AT > histamine = $PGF_{2\alpha}$ > PGE_2). Intense peribronchial oedema and vasoconstriction were observed only in the AT- and $PGF_{2\alpha}$treated animals, but not in the other groups of guinea-pigs. AT, histamine and $PGF_{2\alpha}$ induced perivascular oedema, thickening of the vascular wall and intense emphysema but PGE_2 had no such effects. Neither the untreated nor the native serum treated animals showed any of these histopathological effects.

The results of this study showed that the histopathological effects of AT were mimicked entirely by $PGF_{2\alpha}$, partially by histamine but hardly at all, by PGE_2.

Mepyramine (0.5-4 mg/kg) abolished the effects of histamine but only partially reduced the AT-induced effects. The AT-induced oedema was

particularly resistant to the inhibitory action of mepyramine. On the other hand the anti-inflammatory agents indomethacin and phenylbutazone (0.1-4 mg/kg) abolished the responses to the prostaglandins and effectively inhibited the AT-induced oedema.

It may be concluded from these observations that the histopathological effects of AT are probably mediated through the release of histamine and $PGF_{2\alpha}$, the latter mediator possibly contributing more to the perivascular and peribronchial oedema.

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Release of prostaglandins in the knee joint of the dog during local Shwartzman-like reaction

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The Shwartzmann phenomenon, originally described in the rabbit (Shwartzman, 1928), is a two-stage reaction: the first event consists of a local inflammatory reaction in the skin after an intradermal injection of bacterial endotoxin; the second one consists of a haemorrhagic necrosis of the prepared skin area following an intravenous injection of the endotoxin, 18-24 h after the intradermal injection. Since endotoxin produces an inflammation in the knee joint of the dog (Morgan & Bennett, 1947; Van Arman, Carlson, Kling, Allen & Bondi, 1974) during which prostaglandins (PG) are locally released (Herman & Moncada, 1975), we decided to study the possibility of producing a Shwartzman-like reaction in the dog's knee joint, the release of PG's during its development and the effect of local treatment with indomethacin, which has been shown to inhibit the Shwartzman reaction in rabbit skin (Van Arman, Carlson, Brown & Itkin, 1970).

Mongrel dogs of either sex were anaesthetized with thiopentone (10 mg/kg intravenously). A 'preparatory' injection of endotoxin (E. coli O 111 B4, Difco; 2.5 ng/kg, 9 experiments; 25 ng/kg, 12 experiments) in 0.5 ml of sterile saline was given in one of the knee joint cavities. Symptoms of incapacitation ranging from limping, occasional 3-legged gait to complete 3-legged gait were observed in all animals as described previously (Herman & Moncada, 1975). All animals received an intravenous 'provocative' injection of 250 ng/kg endotoxin 18-24 h later. In 15 out of 21 animals incapacitation occurred which started about 1 h after the injection and lasted for about 3-4 hours. In 12 animals synovial fluid was sampled from the affected joint under light thiopentone anaesthesia and the cavity washed twice with 2 ml of sterile saline. Samples were extracted and bioassayed for PG-like activity (Gilmore, Vane & Wyllie, 1968). In 10 out of 12 animals PG-like activity (range: 4-51 ng PGE_2-equivalents) was detected in the synovial fluid.

In 3 animals indomethacin (200 μg/kg dissolved in phosphate buffer 0.1 M pH 8.9) was injected in the knee joint at the moment of the provocative injection of endotoxin: no symptoms developed in these animals up to 7 h after the injection.

An intravenous injection with endotoxin 24 h after a preparatory injection with concanavalin A (Sigma; 0.1-1 mg/kg, 8 experiments) or lipid A/BSA (supplied by Dr C. Galanos; 5 μg/kg, 2 experiments) gave incapacitation and production of PG's in all animals studied.

These experiments show that a Shwartzman-like reaction can be produced in the knee joint of the dog, during which PG's are released and which can be prevented by local treatment with indomethacin. These findings indicate that PG's may be involved in the local Shwartzman reaction. Furthermore, we think this to be a useful and practical model to study the release of chemical mediators and cells during the local Shwartzman reaction.

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