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ACTIONS OF 16-ARYLOXY ANALOGUES OF PROSTAGLANDIN $F_{2\alpha}$ ON PREPARATIONS RESPONSIVE TO PROSTAGLANDIN ENDOPEROXIDES

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On four prostaglandin endoperoxide-sensitive preparations, namely human platelets *in vitro*, rabbit aortic strip, guinea-pig tracheal chain and pig blood pressure, ICI 79939 and its 11-oxo analogue mimicked the actions of 11,9-(epoxymethano) prostaglandin H_2, and were considerably more active than either prostaglandin $F_{2\alpha}$ or ICI 81008. It is suggested that this activity of ICI 79939 may contribute to its toxicity in experimental animals.

Introduction It has been shown that certain aryloxy analogues of prostaglandin $F_{2\alpha}$ are considerably more potent than prostaglandin $F_{2\alpha}$ itself as luteolytic agents in several animal species (Dukes, Russell & Walpole, 1974; Crossley, 1975). However, one of these substances, ICI 79939, is much more toxic than prostaglandin $F_{2\alpha}$ and other numbers of the series (e.g. ICI 81008) (Dukes *et al.*, 1974). (ICI 79939 is *rac* 17,18,19,20-tetranor-16-*p*-fluorophenoxy prostaglandin $F_{2\alpha}$ and ICI 81008 is *rac* 17,18,19,20-tetranor-16-*m*-trifluoromethylphenoxy prostaglandin $F_{2\alpha}$). Since the prostaglandin endoperoxides, prostaglandins G_2 and H_2, and thromboxane A_2 exhibit a number of potentially noxious actions, for example bronchoconstriction and platelet aggregation (Hamberg, Svensson, Wakabayashi & Samuelsson, 1973; Hamberg, Svensson, Hedqvist, Strandberg & Samuelsson, 1976) it was decided to investigate the effects of ICI 79939 on several preparations sensitive to endoperoxides/thromboxanes. The 11,9-(epoxymethano) analogue of prostaglandin H_2 (Upjohn 46619) was used as the standard compound in place of either prostaglandin G_2 or H_2 because of its greater stability (Bundy, 1975). ICI 81008 and the 11-oxo (prostaglandin D-type) analogue of ICI 79939 were also included in the study.

Methods

Platelet aggregation Human platelet-rich plasma (PRP) was prepared by centrifugation ($200 \times g$ for 15 min) of freshly collected, citrated venous blood. Aggregation of platelets was measured by the optical method (Born, 1962). The cuvette contents, 1.0 ml of PRP, 0.9 ml of 0.9% w/v NaCl solution and 0.1 ml of drug solution, were stirred with a Teflon-coated rod at 1000 rev/min and maintained at 37°C.

Rabbit aortic strip Aortae from young male rabbits were removed immediately after they had been killed. A spiral strip, 3 mm wide, was cut and suspended in a 10 ml organ bath containing Krebs-Henseleit solution (NaCl 118, KCl 5.4, $MgSO_4$ 1.0, $CaCl_2$ 2.5, NaH_2PO_4 1.2, $NaHCO_3$ 25, dextrose 10 mmol/l), gassed with 95% O_2 and 5% CO_2 and maintained at 37°C. Tension changes were recorded with a Grass FTO3 force-displacement transducer linked to a Grass Polygraph.

Guinea-pig tracheal chain Tracheae were removed from guinea-pigs immediately after they had been killed. Five or six rings were tied together and suspended in a 10 ml organ bath containing Krebs-Henseleit solution (composition as above), gassed with 95% O_2 and 5% CO_2 and maintained at 37°C. Isotonic contractions (load 0.3 g) were recorded with a Washington Type II lever transducer. Prostaglandins were left in contact with the tissue for 3 min and a 45 min dose cycle was used to prevent tachyphylaxis.

Pig blood pressure Pigs, weighing between 12.5 and 20 kg, were anaesthetized with pentobarbitone sodium, 40 mg/kg, injected intravenously. Blood pressure was recorded from a carotid artery. Drugs were injected into the descending aorta through a catheter introduced into the left femoral artery.

Compounds The 11-oxo derivative of ICI 79939 was prepared from ICI 79939 by the method of Nishizawa, Miller, Gorman, Bundy, Svensson & Hamberg (1975).

Results The actions of the three aryloxy analogues and prostaglandin $F_{2\alpha}$ were compared with those of 11,9-(epoxymethano) prostaglandin H_2 on human platelets *in vitro*, the rabbit aortic strip, the guinea-pig tracheal chain and the pig BP. Equipotent molar ratios calculated from the respective log dose-response curves are given in Table 1.

11,9-(Epoxymethano) prostaglandin H_2 produced a rapid and reversible aggregation of human platelets at concentrations of 50 to 200 ng/ml. Irreversible aggregation was always produced with 0.5 to 1.0 μg/ml. ICI 79939 and its 11-oxo analogue mimicked these effects whereas prostaglandin $F_{2\alpha}$ and

ICI 81008 did not. The aggregatory responses were not inhibited by indomethacin at 10^{-5} M, a concentration which completely abolished the effect of arachidonic acid.

All the compounds studied contracted the rabbit aorta and the guinea-pig tracheal chain preparations, although ICI 79939 and its 11-oxo analogue were considerably more active than ICI 81008. The actions on the aorta were unaffected by the presence of indomethacin and a combination of receptor antagonists, atropine, mepyramine, methysergide, phenoxybenzamine and propranolol, in the bathing fluid.

In the anaesthetized pig, the injection of 11,9-(epoxymethano) prostaglandin H_2 into the thoracic aorta produced an immediate rise in systemic arterial blood pressure. In addition, on the blood-perfused hind limb the endoperoxide analogue (0.1–2.5 μg) produced a rise in perfusion pressure on close intra-arterial injection. These effects were mimicked by ICI 79939 and equipotent molar ratios were similar to those found with the previously described preparations. The natural enantiomer of ICI 79939 (ICI 87907) was 120 times more active than its mirror image isomer (ICI 87904) on the pig BP.

Discussion On preparations where prostaglandin $F_{2\alpha}$ is more potent than prostaglandin E_2 and the endoperoxides, for example, longitudinal muscle of the rabbit jejunum (contraction) and the rabbit oviduct *in vivo* (rise in intraluminal pressure), both ICI 79939 and ICI 81008 are between 2 and 5 times less active than prostaglandin $F_{2\alpha}$ (P.J. Welburn and R.L. Jones, unpublished observations). However, as reported here, the activity of ICI 79939 on prostaglandin endoperoxide-sensitive preparations is very much higher than that of either prostaglandin $F_{2\alpha}$ or ICI 81008. Furthermore, when the same substitution (*p*-fluorophenoxymethyl for the terminal *n*-pentyl group) is applied to prostaglandin D_2 then inhibitory activity on platelet aggregation (Smith, Silver, Ingerman & Kocsis, 1974; Nishizawa *et al.*, 1975; these investigations) is replaced by pronounced aggregatory activity.

These differences in endoperoxide-like activity may account to some extent for the much higher toxicity of ICI 79939 compared with ICI 81008 in experimental animals.

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Table 1 Activities of 16-aryloxy prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) analogues on endoperoxide-sensitive systems

Preparation and response	*Threshold dose of 11,9-(epoxymethano) PGH_2*	*Equipotent molar ratios: 11,9-(epoxymethano) PGH_2 = 1.0*			
	ng/ml or ng/kg	*ICI 79939*	*11-oxo ICI 79939*	*$PGF_{2\alpha}$*	*ICI 81008*
Human platelets: aggregation	50–100	2.2	7.2	No effect at 50 μg/ml	No effect at 50 μg/ml
Rabbit aortic strip: contraction	0.5–2.0	3.1	9.3	132	2210
Guinea-pig tracheal chain: contraction	2.0–20	1.2	1.5	133	335
Pig BP: pressor	20–30	3.8	4.5	293	1130

Each value is the mean of at least three determinations.

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