J. Pharm. Pharmac., 1976, **28**, 613–616 *Received April* 29, 1976

Effects of animal maturity on smooth muscle and blood pressure responses to prostaglandins E_2 and $F_{2\alpha}$

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The effects of prostaglandins E_2 (PGE_2) and $F_{2\alpha}$ ($PGF_{2\alpha}$) on muscle strips from mature and immature rats and guinea-pigs and on rat blood pressure were investigated in the presence of atropine. The colon and stomach strips from immature rats were equally responsive to PGE_2 and $PGF_{2\alpha}$ whereas mature colons were significantly more sensitive to $PGF_{2\alpha}$ and mature stomach strips significantly more sensitive to PGE_2. On the ileum from immature guinea-pigs the maximum responses to PGE_2 and $PGF_{2\alpha}$ were 16 and 8% of the histamine maximum respectively. The corresponding figures on the mature ileum were 86 and 75%. Whereas PGE_2 was only twice as active as $PGF_{2\alpha}$ on immature ilea, it was ten times more active than $PGF_{2\alpha}$ on mature muscles. On blood pressure $PGF_{2\alpha}$ and PGE_2 were both hypotensive in immature rats whereas PGE_2 was hypotensive and $PGF_{2\alpha}$ hypertensive in mature rats. The results suggest that as the animal gets older, receptors for prostaglandins became increasingly differentiated.

Smooth muscle strips from colon and stomach of rats are sensitive to low concentrations of prostaglandins. Compared to F-type prostaglandins, E compounds have been shown to be weakly active on colon muscles while the stomach muscle is less sensitive to PGFs (Ferreira, Moncada & Vane, 1973). In our laboratory, we found large variations in the relative sensitivities of these muscles to PGE_2 and $PGF_{2\alpha}$. In this report, results are presented which show that the relative potencies of PGE_2 and $PGF_{2\alpha}$ in contracting rat and guinea-pig smooth muscles and their effects on rat blood pressure vary with animal maturity.

METHODS

Rat colon and rat stomach strip

Inbred albino rats of either sex were grouped into mature rats (350–400 g) and immature rats (35–100 g). The animals were starved overnight but allowed water. Colons and stomach strips were isolated and set up as described by Vane (Vane, 1957; Regoli & Vane, 1964). The strips were superfused in series with 5 ml min^{-1} aerated Tyrode solution at 37° using a Watson-Marlow flow inducer (MHRE 22). The Tyrode solution was of the following composition (g $litre^{-1}$): NaCl 8·0, KCl 0·2, $CaCl_2$ 0·05, Mg Cl_2 0·05, $NaHCO_3$ 1·0, NaH_2PO_4 0·05, Glucose 1·0. Atropine (10^{-6} g ml^{-1}), propranolol (10^{-7} g ml^{-1}) and phentolamine (10^{-6} g ml^{-1}) were added to minimize effects due to acetylcholine and catecholamine release. Contractions of the longitudinal muscles were recorded on smoked paper by auxotonic (Paton, 1957) frontal writing levers with an initial load of 2·0 g. The tissues were allowed 1 h to equilibrate before PGs were added. PGE_2 or $PGF_{2\alpha}$ (2·5, 5, 10, 20 and 40 ng ml^{-1}) were infused for 30 s at intervals of 5 min or as soon as the tissue had relaxed to the base-line.

Guinea-pig ileum

8 adult (500–600 g), and 7 immature (130–150 g) guinea-pigs were used. Ileal segments about 2 cm long were taken 5 cm away from the caecum. The muscle was suspended in aerated Tyrode solution containing atropine 10^{-6} g ml^{-1} at 37° in a 15 ml jacketed organ bath. Contractions of the longitudinal muscle were recorded on smoked paper with a frontal writing lever (magnification ×6 and a load of 1 g).

Rat blood pressure

Rats were anaesthetized with urethane (1·5 g kg^{-1}, i.p.). Mean arterial blood pressure was recorded from the common carotid artery with a Bell & Howell pressure transducer (Type 4–327–L223) on a Model M19 Devices Recorder. Drugs were injected via the femoral vein in volumes not exceeding 0·1 ml and washed in with 0·2 ml of saline. All animals received bretylium 0·5 mg kg^{-1} and atropine 1 mg kg^{-1} (i.p.) 30 min before.

RESULTS

Rat colon and rat stomach

The relative sensitivities of colon and stomach preparations from mature and immature rats are

shown in dose-response curves in Fig. 1. Since previous reports have shown that the colon is more sensitive to $PGF_{2\alpha}$ than to PGE_2, the response to each dose of PG on the colon was expressed as a percentage of the response to the highest concentration of $PGF_{2\alpha}$ (40 ng ml^{-1}). Similarly, all responses of the stomach strip were expressed as a percentage of the response to 40 ng ml^{-1} PGE_2. The tissues from mature rats exhibited marked differential sensitivities to PGE_2 and $PGF_{2\alpha}$, the colon being more sensitive to $PGF_{2\alpha}$ than PGE_2 (Student's *t*-test $P < 0{\cdot}01$); the responses to PGE_2 were approximately half those to $PGE_{2\alpha}$ at all dose levels. In stomach strips from mature animals responses to PGE_2 were greater at all dose levels than those to $PGF_{2\alpha}$ ($P < 0{\cdot}001$). At lower doses (2·5, 5 and 10 ng ml^{-1}) mature stomachs were 3–4 times more sensitive to PGE_2 than $PGF_{2\alpha}$.

Muscle strips from immature rats on the other hand showed similar sensitivities to the PGs ($P > 0{\cdot}2$). The age-dependent differences were due to decreased sensitivities of rat colon to PGE_2 and rat stomach to $PGF_{2\alpha}$. Their sensitivities to $PGF_{2\alpha}$ and PGE_2 respectively were similar in tissues from mature and immature animals.

Guinea-pig ileum

Doses of PG were given at 5 min intervals with a contact time of 60 s. Tachyphylaxis occurred with shorter time intervals. PG dose-response curves were constructed by expressing contractions as a percentage of the maximum response to histamine (Fig. 2). On the ileum from immature guinea-pigs, PGE_2 and $PGF_{2\alpha}$ were weakly active. The maximum responses to PGE_2 and $PGF_{2\alpha}$ were 16 ± 6 and 8 ±2% (mean ± s.e.m., n = 7) of the maximum response to histamine respectively. Although PGE_2 produced higher contractions than $PGF_{2\alpha}$ on immature ilea, the threshold dose of about 30 ng ml^{-1} was the same for both PGs. On ilea from mature guinea-pigs, the maximum responses to PGE_2 and $PGF_{2\alpha}$ were 86 ± 1 and 76 ± 6% (mean ± s.e.m., n = 8) of the histamine maximum respectively. Although PGE_2 caused only a slightly greater maximum than $PGF_{2\alpha}$, the threshold doses for minimum stimulations were different (3 ng ml^{-1} for PGE_2 and 30 ng ml^{-1} for $PGF_{2\alpha}$). The mature ilea were slightly more sensitive to histamine than the immature ilea but the maximum responses to histamine appeared to be similar.

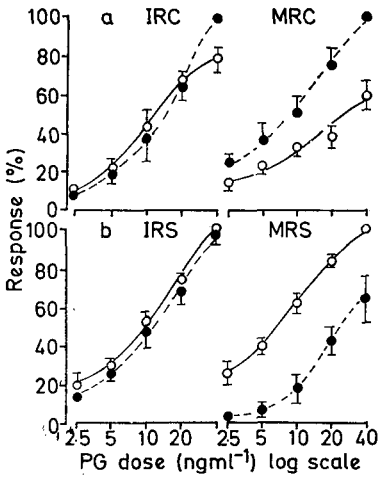

FIG. 1. Dose response curves to and a-$PGF_{2\alpha}$, ●– – –● and b-PGE_2, ○——○ on colons from immature rats (IRC) and mature rats (MRC), and on stomach strips from immature rats (IRS) and mature rats (MRS). Each point is a mean of measurements taken from at least six separate muscle preparations. The vertical bars represent standard errors of the mean.

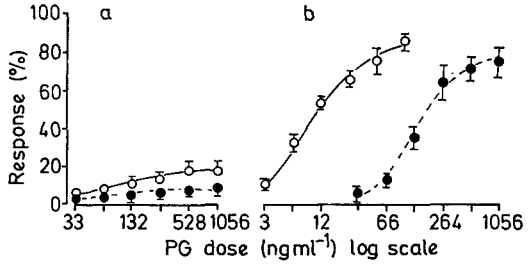

FIG. 2. Dose response curves to PGE_2, ○——○ and $PGF_{2\alpha}$ ●– – –● in ileal muscles taken from a-immature and b-mature guinea-pigs. The response to each dose of the PG was expressed as a percentage of the maximum response of the tissue to histamine. Each point is a mean of measurements taken in muscles from 7 immature and 8 mature guinea-pigs. The vertical bars indicate standard errors of the mean. The Tyrode solution contained atropine (10^{-6} g ml^{-1}).

Rat blood pressure

PGE_2 lowered arterial blood pressure in both adult and immature rats. In each of 7 adult rats $PGF_{2\alpha}$ raised blood pressure at all dose levels whereas the predominant effect of $PGF_{2\alpha}$ in immature rats was a fall, which was not always clearly related to the dose of the prostaglandin. The hypotensive effect of $PGF_{2\alpha}$ in immature rats was more complex than that of PGE_2. Following the injection of $PGF_{2\alpha}$ blood pressure fell sharply but recovered in about 30 s. This was followed by a hypotension phase lasting 5–10 min (Fig. 3). The blood pressure changes in immature rats shown against $PGF_{2\alpha}$ in Table 1 refer to the maximum fall in blood pressure caused by each dose in the second hypotensive phase.

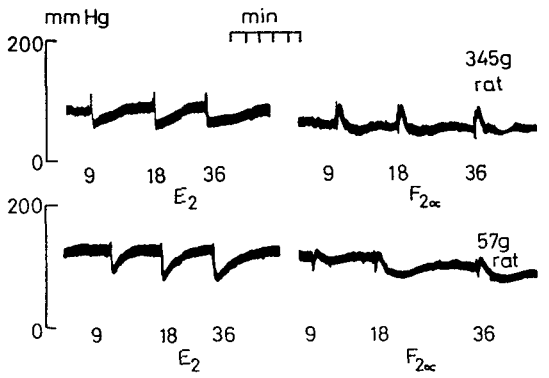

FIG. 3. Effects of prostaglandins E_2 and $F_{2\alpha}$ on the blood pressure of an immature and a mature rat. The doses under the responses are $\mu g\ kg^{-1}$ body weight injected intravenously.

Table 1. *Blood pressure changes induced by PGE_2 and $PGF_{2\alpha}$ in immature and mature rats.*

PG	Dose	Changes in blood pressure (mm Hg ± s.e.m) in Immature rats Body wt (50–100 g) (n = 7)	Mature rats Body wt (300–400 g) (n = 7)
	$\mu g\ kg^{-1}$		
E_2	4·5	−19 ± 3	−10 ± 2
	9	−30 ± 3	−14 ± 3
	18	−40 ± 2	−17 ± 4
	36	−47 ± 3	−23 ± 5
$F_{2\alpha}$	4·5	− 8 ± 2	+15 ± 3
	9	−13 ± 4	+23 ± 5
	18	−27 ± 6	+28 ± 4
	36	−20 ± 5	+38 ± 7

At all dose levels, PGE_2 caused a greater fall than $PGF_{2\alpha}$ in blood pressure of immature rats ($P < 0{\cdot}01$).

DISCUSSION

The results show that there are marked age-dependent differences in smooth muscle and rat blood pressure responses to PGE_2 and $PGF_{2\alpha}$. In the longitudinal muscle of guinea-pig ileum, PG E and F compounds act on both cholinergic nerves and muscle cells (Bennett, Eley & Stockley, 1975). In the present experiments, responses were elicited in the presence of atropine, and were presumably due to direct stimulation of the muscle. The lower maxima of the PG dose-response curves (less than 20% of the histamine maxima) in immature muscles compared with maxima of about 80% in mature muscles might indicate a higher PG receptor population in mature muscles. Also, the threshold concentration of PGE_2 for minimal stimulation was ten times lower in mature ileum. Bennett & others (1975) proposed that in the longitudinal muscle of the guinea-pig ileum, PGE_2 and $PGF_{2\alpha}$ may act at a common receptor site, since the pA_2 values of the PG antagonist SC-19220 [1-acetyl-2-(8-chloro-10,11-dihydrobenz[b,f][1,4]-oxazepine-10-carbonyl)-hydrazine] against the compounds were similar (5·5 and 5·6) (Bennett & Posner, 1971). Their results involved the action of PGs at neural and muscle sites whereas in the present experiments, only muscle sites were studied. However, the difference in relative potencies of PGE_2 and $PGF_{2\alpha}$ in mature and immature muscles may indicate a change in the receptors during maturation. Another factor might be a higher PG receptor reserve in mature muscles. Although PGE_2 was less than twice as potent as $PGF_{2\alpha}$ in immature guinea-pig ilea, it was ten times more active than $PGF_{2\alpha}$ on mature muscles, indicating an increased sensitivity of the 'mature' receptors to PGE_2. There was also indication of receptor changes with age in rat muscles. Whereas the two prostaglandins were equally active on colon and stomach muscles from immature rats, there were significant differencies in their relative activities on muscles from mature rats.

On blood pressure, PGE_2 was hypotensive in mature and immature rats, but whereas $PGF_{2\alpha}$ was hypertensive in adult rats, it was predominantly hypotensive in immature rats. These effects can also be attributed to a direct action of the PGs on vascular smooth muscle since reflex and indirect mechanisms should be minimal in animals treated with atropine and bretylium. The age-related differences in blood pressure responses to the PGs was unlikely to reflect differences in resting blood pressure, because although the resting mean blood pressure in immature rats (116 ± 4, n = 7) tended to be higher than that of mature rats (109 ± 5, n = 7) the difference was not significant ($P > 0{\cdot}1$). Here again, the differences in blood pressure responses to PGE_2 and $PGF_{2\alpha}$ appear to be related to age-dependent changes in receptor characteristics.

The present results suggest that muscle receptor sites for PGE_2 and $PGF_{2\alpha}$ may be different at least at some time in the animal's life. Changes in receptor population and characteristics with animal maturity may be a common property of drug receptors since Botting (1975) has recently demonstrated a similar phenomenon for histamine H_1-receptors in the rabbit ileum. Wood, Saverymuttu & Morton (1975) have also shown age-dependent differences in the effect of PGs on guinea-pig gall bladder fluid transport.

Acknowledgements

I thank Dr J. E. Pike (Upjohn, Kalamazoo, U.S.A.) for gifts of PGs and Miss F. Oladitan and Mr J. Nwaubani for technical assistance. The work was supported by grants from the University of Ibadan Senate Research Fund and the Medical Research Council of Nigeria.

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