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Effects of prostaglandin E_2 on fast and slow components of the response of the rat vas deferens to field stimulation

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1 The response of the rat vas deferens to field stimulation with single pulses is biphasic. The first, rapid component of the response is dominant at the prostatic end and is thought by some to be mediated by non-adrenergic, non-cholinergic neurotransmission. The second, slower component predominates at the epididymal end and has the usual properties associated with adrenergic responses.

2 Prostaglandin E_2 (PGE_2, 10^{-8} to 10^{-5} mol/l) inhibited the 'fast component' responses elicited by field stimulation of the prostatic vas from reserpine-treated rats. The prostaglandin was more potent in the presence of 1.25 than 2.5 mmol/l Ca^{2+}.

3 PGE_2 (10^{-7} to 10^{-5} mol/l) had similar effects on prostatic preparations from normal rats, but was less active on these.

4 PGE_2 (10^{-9} to 10^{-5} mol/l) did not inhibit the 'slow component' responses resulting from field stimulation of the epididymal vas but rather, at the two highest concentrations used (5×10^{-6} and 10^{-5} mol/l), potentiated them. These high concentrations of the prostaglandin also potentiated the contractions of the epididymal vas elicited by exogenous noradrenaline.

5 These effects of PGE_2 on the two components of the response of the rat vas deferens are not as might be expected from its inhibitory effects on the 'typical' adrenergic neurotransmission in several other sympathetically-innervated tissues.

Introduction

The mechanical response of the rat vas deferens to hypogastric nerve or field stimulation is biphasic (Swedin, 1971; Anton, Duncan & McGrath, 1977; McGrath, 1978). The two components underlying the two phases of the response can best be revealed when responses are elicited by single electrical stimuli rather than by trains of pulses (McGrath, 1978). The first, rapid component of the response to a single stimulus reaches a peak at about 250 ms, and is followed by a second, slower component peaking at about 650 ms. The relative contribution of these two components to the overall response varies between the different ends of the vas. The first component is the dominant response at the prostatic end, whereas the slower second component is dominant at the epididymal end (McGrath, 1978; Brown, McGrath & Summers, 1979; MacDonald & McGrath, 1980a, b).

There is general agreement that the second component of the response is a typical adrenergic response mediated by the action of noradrenaline on α-adrenoceptors, since it can be eliminated by reserpine pretreatment or by postjunctional α-adrenoceptor antagonists (MacDonald & McGrath, 1980b). In contrast, the first component is apparently resistant to reserpine-treatment, is not reduced by α-adrenoceptor antagonists, and also survives apparent destruction of adrenergic terminals with 6-hydroxydopamine (Anton *et al.*, 1977; McGrath, 1978; Booth, Connell, Docherty & McGrath, 1978; Brown *et al.*, 1979). Thus, some authors are of the opinion that the first component is due to a separate type of non-adrenergic junction (Booth *et al.*, 1978; Brown *et al.*, 1979) while others favour the view that both components of the effector response are adrenergic, but the first is relatively resistant to postjunctional blockade and the effects of reserpine (see Sjöstrand, 1981).

Prostaglandins of the E series have been shown to inhibit adrenergic neurotransmission in a variety of tissues (reviewed by Hedqvist, 1977), and have also been reported to inhibit neuroeffector transmission in the rat vas deferens. Thus, prostaglandin E_1 (PGE_1) and prostaglandin E_2 (PGE_2) reduced longitudinal contractile responses of the vas to field stimulation (Hedqvist & von Euler, 1972). However, in this study, responses were elicited by trains of stimuli rather than single pulses. Under these condi-

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tions even the first, rapid phase of the response contains elements from both components of the response seen with single stimuli (McGrath, 1978). Tomlinson (1980), using a method which measured predominantly contractions of the circular smooth muscle of the vas, found that PGE_1 inhibited responses to field stimulation, although there was no clear concentration-effect relationship. Again, responses were elicited by trains of stimuli. Ambache, Dunk, Verney & Zar (1972) observed little or no effect of PGE_2 on the longitudinal contractions of the rat vas deferens induced by field stimulation with single pulses, but at that time it was not recognized that such responses (i.e. those evoked by single pulses) comprise two components, separable by time-course. More striking inhibitory effects of PGE_1 and PGE_2 have been reported on responses to field or hypogastric nerve stimulation in the guinea-pig vas deferens (Ambache & Zar, 1970; Baum & Shropshire, 1971; Swedin, 1971; Hedqvist & von Euler, 1972), in which similar mechanisms may be involved in neurotransmission as in the rat. However, all these studies were carried out using trains of stimuli, precluding any clear distinction between the effects of the prostaglandins on the two components contributing to the total response. Thus, there is little information about the effects of prostaglandins on the individual components of the response of the rat or guinea-pig vas deferens to field or hypogastric nerve stimulation. By the use, in different experiments, of epididymal and prostatic portions of the rat vas deferens, pretreatment of animals with reserpine as appropriate, and field stimulation with single pulses, we have sought to examine separately the effects of PGE_2 on the slow (adrenergic) and fast components of the response of this tissue to intramural nerve stimulation.

Methods

Vasa deferentia were dissected from freshly-killed adult Wistar rats. Lengths of either the epididymal or the prostatic end, representing approximately one third of the total length of the vas, were suspended in an organ bath, of volume 12 or 13 ml, between a pair of parallel silver: silver chloride plate electrodes. The organ bath contained Krebs-Henseleit solution (referred to subsequently as Krebs solution) of the following composition (mmol/l): NaCl 118.8, KCl 4.15, $NaHCO_3$ 25.5, $MgSO_4$ 1.2, KH_2PO_4 1.23, glucose 11.1, $CaCl_2$ 1.25 or 2.5. The solution was maintained at a temperature of 37°C and gassed with 95% O_2 and 5% CO_2. Tension was measured with a Devices 2STO2 strain gauge and recorded on a Devices M4 polygraph. The initial resting tension was set at 0.5 g.

In two series of experiments with the prostatic end of the vas, the rats were injected with reserpine, 3 mg/kg intraperitoneally, 18 h before use. This treatment has been reported to eliminate the slow component of the response to field stimulation (McGrath, 1978; MacDonald & McGrath, 1980a, b), facilitating measurement of the fast component.

After an initial equilibration period of approximately 1 h, contractions were elicited by single pulses of 1 ms duration and a supramaximal voltage, and recorded at a chart speed of 5 or 10 mms^{-1}. The pulses were applied at intervals of 5 min, to avoid possible modification of the responses by endogenous feedback mechanisms (McGrath, 1978). Routinely, the organ bath was emptied and refilled after every second stimulus, i.e. at intervals of 10 min. When at least four responses of similar magnitude had been obtained, PGE_2 was added to the organ bath and four more responses were obtained (the organ bath being washed out after the second response as before, and the dose of PGE_2 replenished). In this way, the means of four responses before (control) and during the presence of PGE_2 were obtained, enabling calculation of the percentage change in response in the presence of the prostaglandin. This sequence of 'control' and 'test' responses was repeated, using different concentrations of PGE_2. Thus, in each experiment, the effects of a range of concentrations of the prostaglandin were studied. The concentration ranges used, in different series of experiments, were 10^{-9} to 10^{-7}, 10^{-8} to 10^{-6}, 10^{-7} to 10^{-5} or 2×10^{-7} to 10^{-5} mol/l.

The influence of PGE_2 on the responses of the (epididymal) vas to noradrenaline was determined in a similar manner, except that contractions were evoked by adding a sub-maximal concentration of noradrenaline to the organ bath rather than by electrical stimulation. A dose cycle of 6.5 min was used, with a noradrenaline contact time of 1.5 min. After at least two consistent 'control' responses had been obtained, a dose of PGE_2 was added (immediately after wash-out of the noradrenaline) and two more responses obtained. The dose of PGE_2 was replenished after the bath had been washed out following the first of these responses. Again, this sequence was repeated using different concentrations of the prostaglandin, from 2×10^{-7} to 10^{-5} mol/l.

In some experiments nifedipine (3×10^{-6} or 6×10^{-6} mol/l) was used to abolish selectively the fast component of the response to field stimulation (French & Scott, 1981), enabling examination of the slow component alone.

Tests of statistical significance were carried out using Student's *t* test.

Drugs

Prostaglandin E_2 was purchased from Sigma Chemi-

cal Co.; solutions were prepared in Krebs solution containing 1.25 or 2.5 mmol/l Ca^{2+} as appropriate to the experiment. Noradrenaline bitartrate (Sigma) was dissolved in 0.9% w/v NaCl solution (saline). Nifedipine (Bayer Ltd) was dissolved in Cremophor EL (Sigma) and diluted with saline immediately before use; all operations with this compound were carried out under sodium light. Reserpine (Sigma) was dissolved in 1% v/v aqueous acetic acid to give a solution containing 1.5 mg/ml; this solution was rendered less acidic by the addition of 60 µl 2M NaOH per 5 ml of solution, immediately before injection. Fresh solutions of all these drugs were prepared on each day they were used. Tetrodotoxin (Sigma) was stored as a stock solution of 10^{-4} mol/l in aqueous citrate buffer, pH 4.8, and diluted with saline immediately before use.

Results

Responses of the rat vas deferens to field stimulation under the conditions of our experiments were abolished by tetrodotoxin (10^{-7} mol/l), confirming that they were neurally mediated. This was established both in preparations bathed in Krebs solution containing 1.25 and 2.5 mmol/l Ca^{2+}.

Effect of prostaglandin E_2 on the response of the prostatic vas to field stimulation: preparations from reserpine-treated rats

The response of the prostatic vas from reserpine-treated rats to single stimuli consisted of a monophasic fast contraction, peaking at approximately 250 ms, as described previously (MacDonald & McGrath, 1980a). In preparations bathed in Krebs solution containing 1.25 mmol/l Ca^{2+}, the mean tension developed initially was 0.21 ± 0.03 g (mean ± s.e.mean, $n = 10$). PGE_2 caused a dose-related inhibition of the response (Figure 1). This inhibition was significant ($P < 0.001$) at each concentration of PGE_2 used (10^{-8} to 10^{-6} mol/l), and the highest concentration caused approximately 75% inhibition.

This series of experiments was repeated at a Ca^{2+} concentration of 2.5 mmol/l. The twitches evoked by field stimulation were stronger under these conditions, the mean tension developed initially being 1.09 ± 0.07 g (mean ± s.e.mean, $n = 8$). Once again PGE_2 reduced the responses ($P < 0.05$ at a concentration of 10^{-7} mol/l, $P < 0.01$ at all other concentrations used, from 2×10^{-7} to 10^{-5} mol/l) but the prostaglandin was considerably less potent at this higher Ca^{2+} concentration. This was evident from a substantial shift of the concentration-effect curve to the right (Figure 1).

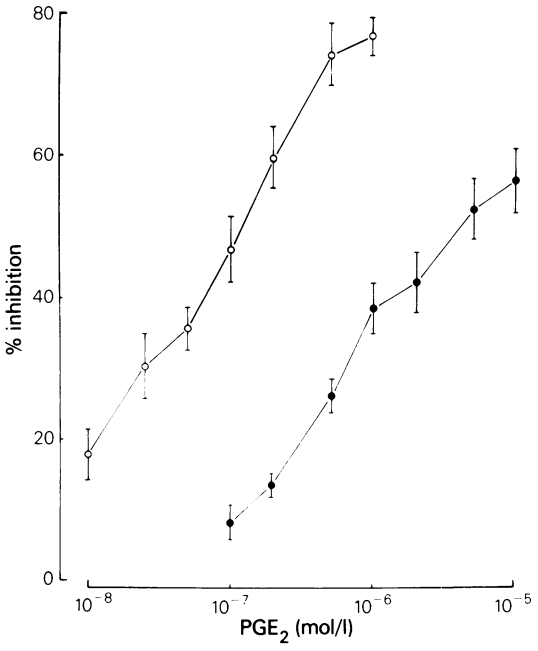

Figure 1 Inhibition by prostaglandin E_2 (PGE_2) of responses of the prostatic end of the vas deferens to field stimulation with single pulses. Preparations from reserpine-treated rats, bathed in Krebs solution containing 1.25 (○) or 2.5 (●) mmol/l Ca^{2+}. Mean results from 10 (○) or 8 (●) preparations: vertical bars represent s.e.mean.

Effects of prostaglandin E_2 on the response of the prostatic vas to field stimulation: preparations from normal rats

In Krebs solution containing 1.25 mmol/l Ca^{2+}, field stimulation of the prostatic end of the vas from non-reserpine-treated (normal) rats again evoked a fast response, peaking at 250 ms. The tension developed initially was 0.37 ± 0.04 g (mean ± s.e.mean, $n = 8$); this was significantly greater ($P < 0.01$) than in the preparations from the reserpine-treated rats. Although the response often appeared to consist only of one component, treatment with nifedipine (3×10^{-6} mol/l) abolished this fast response but revealed a second, much smaller and slower component, peaking at approximately 650 ms. Under these conditions, the magnitude of this slow component varied from 2.4 to 18.2% (mean 10.3%) of the 'fast' component measured before the addition of nifedipine (determined in 6 preparations). However, the time-course of the slow component was such that its magnitude at 250 ms was only in the region of 5% of its peak (650 ms) value. Thus, it was considered that the presence of this slow component would have

contributed very little to the measured 'fast' response peaking at 250 ms in the absence of nifedipine.

As before, PGE_2 inhibited the response of the prostatic vas to field stimulation ($P<0.05$ at a concentration of 10^{-7} mol/l, $P<0.01$ at all other concentrations used, from 2×10^{-7} to 10^{-5} mol/l; Figure 2). However, it appeared to be considerably less potent than in the reserpine-treated rats, as can be seen from a comparison of Figure 2 with Figure 1. Furthermore, the maximum inhibition obtained was only about 45% (cf. the 75% inhibition obtained with the preparations from reserpine-treated rats).

It seemed possible that the lower potency of PGE_2 on the vasa from normal rats was related to the greater 'control' tensions developed by these preparations. In an attempt to gain information about this, an analysis was carried out to test the correlation between the percentage inhibition seen in each experiment at one particular concentration of PGE_2 (10^{-6} mol/l) and the tension developed in the control responses just before addition of the prostaglandin. There was no significant correlation between 'control' tension and percentage inhibition, in preparations from either reserpine-treated or normal rats ($r=-0.005$ and -0.067 respectively).

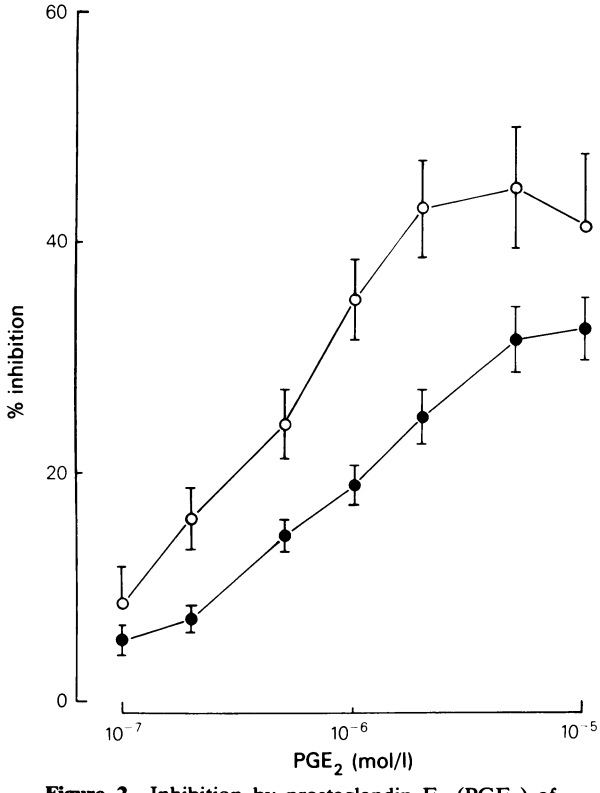

Figure 2 Inhibition by prostaglandin E_2 (PGE_2) of responses of the prostatic end of the vas deferens to field stimulation with single pulses. Preparations from non-reserpine-treated (normal) rats bathed in Krebs solution containing 1.25 (○) or 2.5 (●) mmol/l Ca^{2+}. Mean results from 8 preparations in each case: vertical bars represent s.e.mean.

The experiments with preparations from normal rats were repeated in Krebs solution containing 2.5 mmol/l Ca^{2+}. Under these conditions, the mean tension developed initially (1.17 ± 0.08 g, mean ± s.e.mean, $n=8$) was greater than at the lower Ca^{2+} concentration, but not significantly different ($P>0.4$) from that developed by the preparations from reserpine-treated rats in 2.5 mmol/l Ca^{2+}. However, at this higher level of Ca^{2+} responses of the prostatic vas from normal rats usually showed a small inflection as they decayed from their 250 ms peak, indicative of the presence of the second, slow component. Experiments with nifedipine (3×10^{-6} or 6×10^{-6} mol/l) confirmed that this second component was more marked at 2.5 than at 1.25 mmol/l Ca^{2+}. Its peak magnitude (at 650 ms) ranged from 7.6 to 44.4% (mean 26.9%) of the response measured at 250 ms before the addition of nifedipine (determined in 7 preparations). Nevertheless, in view of the observation described above that the magnitude of the slow component at 250 ms was only approximately 5% of its peak value at 650 ms, its presence was considered once more to contribute little to the magnitude of the measured 'fast' response peaking at 250 ms in the absence of nifedipine.

In the presence of 2.5 mmol/l Ca^{2+}, PGE_2 again inhibited the response of the prostatic vas to field stimulation ($P<0.01$ at a concentration of 10^{-7} mol/l, $P<0.001$ at all other concentrations used, from 2×10^{-7} to 10^{-5} mol/l), but as before it was less effective than at a Ca^{2+} concentration of 1.25 mmol/l (Figure 2). Comparison of Figure 2 with Figure 1 shows that at the higher Ca^{2+} concentration the prostaglandin again had less effect on the preparations from normal rats than on those from the reserpine-treated animals.

Effect of prostaglandin E_2 on the response of the epididymal vas to field stimulation

The response of the epididymal vas to single stimuli consisted predominantly of the 'slow component' peaking at approximately 650 ms described above. However, a smaller fast component was also present, as indicated by an inflection in the rising phase of the response at 250 ms. This fast component could be abolished by nifedipine (3×10^{-6} mol/l), with little or no effect on the slow component. However, because nifedipine is described as a calcium antagonist (Fleckenstein, 1977) and the effects of PGE_2 on sympathetic neurotransmission in other tissues seem to be related to calcium availability (see Hedqvist,

1977), it seemed possible that it might interfere with any effect of PGE_2 on these 'slow component' responses. Thus, we thought it undesirable to use nifedipine in this particular study.

Inspection of the monophasic fast responses obtained by field stimulation of the prostatic vas from reserpine-treated rats showed that by 650 ms, the fast component had decayed to approximately 30% of its peak (250 ms) value. In view of this, we considered that measurement of the peak value (at 650 ms) of the response of the epididymal vas would give an acceptable measure of the magnitude of the 'slow' component. Although such a measurement would include a contribution from the (smaller) fast component, we thought it preferable to accept this rather than to use nifedipine.

In Krebs solution containing 1.25 mmol/l Ca^{2+}, concentrations of PGE_2 from 2×10^{-7} to 2×10^{-6} mol/l appeared to have little or no effect on this 'slow' component of the response of the vas deferens (Figure 3a). However, in marked contrast to the results obtained for the fast component responses of the prostatic vas, higher concentrations of the prostaglandin (5×10^{-6} and 10^{-5} mol/l) caused a substantial potentiation of the response ($P < 0.001$). PGE_2, at concentrations used in this series of experiments, was also found to increase the resting tone of the epididymal vas slightly (no such effect of the prostaglandin on resting tone was seen in preparations from the prostatic end of the vas).

When these experiments were repeated at the higher Ca^{2+} concentration of 2.5 mmol/l, similar results were obtained except that the potentiations evoked by the high concentrations of PGE_2 were smaller (Figure 3a). As expected, the initial tension developed by preparations bathed in Krebs solution containing 2.5 mmol/l Ca^{2+} was greater than that developed by the preparations bathed in 1.25 mmol/l Ca^{2+} (0.35 ± 0.04 g and 0.14 ± 0.02 g respectively, means ± s.e.mean, $P < 0.01$).

As described above, our measurements of the 'slow component' response of the epididymal vas

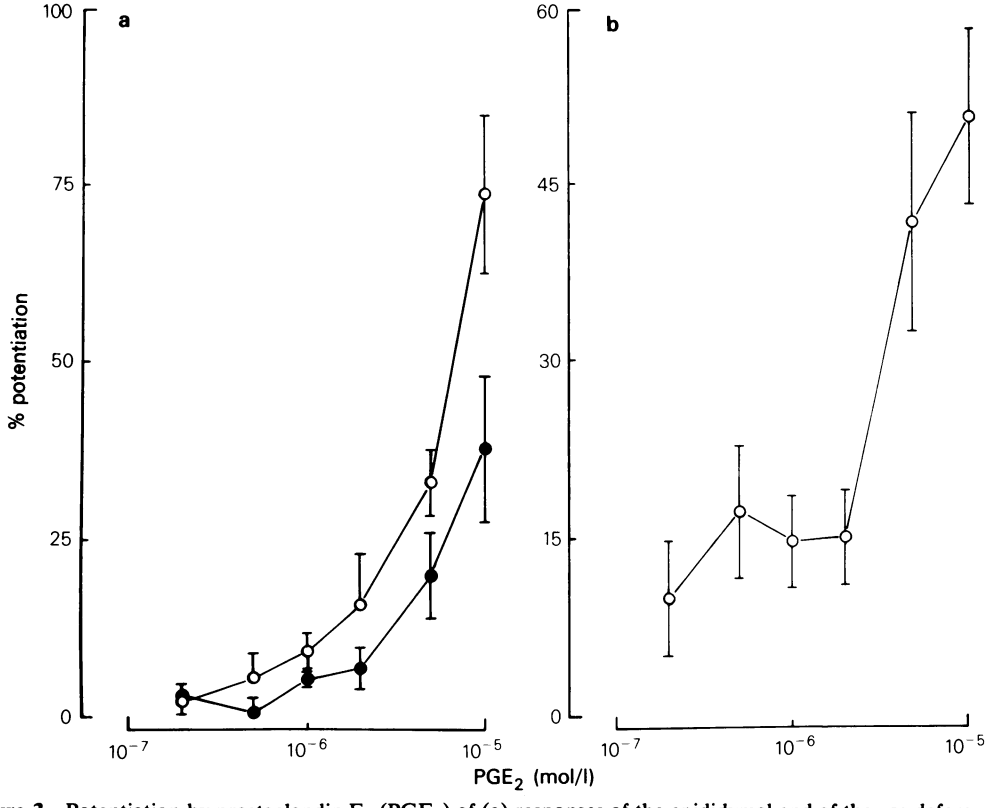

Figure 3 Potentiation by prostaglandin E_2 (PGE_2) of (a) responses of the epididymal end of the vas deferens to field stimulation with single pulses, (b) tonic responses of the epididymal vas to noradrenaline. Preparations from normal rats, bathed in Krebs solution containing 1.25 (○) or 2.5 (●) mmol/l Ca^{2+}. Mean results from 8 preparations in each case: vertical bars represent s.e.mean.

would have included a small contribution from the fast component. Since we had shown PGE_2 to inhibit this fast component, the potentiation of the slow component by the prostaglandin as measured in this series of experiments might be slightly underestimated.

To examine the possibility that low concentrations of PGE_2 had an inhibitory effect on the 'slow component' responses of the epididymal vas, that was masked at higher concentrations by the potentiating effect, a separate series of experiments was carried out using much lower concentrations of the prostaglandin. However, no consistent inhibition or potentiation of the responses was seen at any of 6 concentrations of PGE_2 ranging from 10^{-9} to 10^{-7} mol/l (tested on 6 preparations in the presence of 1.25 mmol/l Ca^{2+}).

Effect of prostaglandin E_2 on the response of the epididymal vas to noradrenaline

These experiments were carried out in Krebs solution containing 1.25 mmol/l Ca^{2+}. Exposure of the epididymal vas to noradrenaline evoked an increase in tone which reached a peak within 90 s. Frequently, small, irregular twitch-like contractions were superimposed on the tonic contraction. A dose of noradrenaline was chosen which gave approximately 30% of the maximum (tonic) response. This dose varied between different preparations; it was either 5×10^{-6} or 10^{-5} mol/l, and in the eight experiments comprising this series the responses thus evoked ranged from 25–36% of the maximum. The effects of PGE_2 on the tonic responses evoked by noradrenaline are shown in Figure 3b. Exposure to PGE_2 at concentrations between 5×10^{-7} and 2×10^{-6} mol/l was followed by an increased response to noradrenaline ($P < 0.05$ at a PGE_2 concentration of 5×10^{-7} mol/l, $P < 0.01$ at PGE_2 concentrations of 10^{-6} and 2×10^{-6} mol/l), but this apparent potentiation was small (about 15%) and did not appear to be dose-related. However, as in the experiments where contractions of the epididymal vas were evoked by field stimulation, higher concentrations of the prostaglandin (5×10^{-6} and 10^{-5} mol/l) caused a marked potentiation of the response ($P < 0.01$ at 5×10^{-6} mol/l, $P < 0.001$ at 10^{-5} mol/l). There was no indication of any continued enhancement of the responses of the epididymal vas to noradrenaline (or to field stimulation) following wash-out of the prostaglandin.

PGE_2 had no obvious effect on the small, twitch-like contractions superimposed on the tonic contractions evoked by the noradrenaline; however, these were very variable and we did not attempt to quantify them.

Discussion

The results of our experiments indicate that PGE_2 inhibits the fast component, but not the slow component, of the response of the rat vas deferens to field stimulation with single pulses. Although there is considerable evidence that the fast component of the response is non-adrenergic in origin, whilst the slow component is adrenergic, this interpretation is by no means universally accepted, and the possibility must also be considered that both components are adrenergic. Therefore, there are a number of possible interpretations of our results.

Assuming, first, that the first component of the response is non-adrenergic, our results suggest that PGE_2 inhibits either the release of, or the response of the tissue to, the non-adrenergic transmitter. In contrast, the prostaglandin appeared to have no effect on the adrenergic transmission, except at high concentrations where facilitation occurred. This facilitation of the adrenergic transmission seems to involve a post-junctional mechanism, because the response of the (epididymal) vas to exogenous noradrenaline was also potentiated.

There are several other examples of apparently non-adrenergic, non-cholinergic neurotransmission in the peripheral nervous system (Gillespie, 1972; Ambache & Killick, 1978; Daniel, 1979; Krell, McCoy & Ridley, 1981) but there have been few reports of the effects of prostaglandins on responses mediated by innervations of this nature. Al Timimi, Bedwani & Stanton (1978) found that PGE_2 reduced the inhibitory responses (relaxations) of the rat anococcygeus muscle induced by field stimulation in the presence of guanethidine, but concluded that this was probably due to an increase in tone evoked by the prostaglandin, rather than a specific effect on the non-adrenergic, non-cholinergic neurotransmission. Burnstock, Cocks, Crowe & Kasakov (1978) found that PGE_1 and PGE_2, as well as $PGF_{2\alpha}$, potentiated the response of the guinea-pig urinary bladder to non-adrenergic, non-cholinergic nerve stimulation, and Husted, Sjoegren & Andersson (1980) reported similar effects of PGE_2 and $PGF_{2\alpha}$ on the rabbit detrusor.

Alternatively, if one takes the view that both fast and slow components of the response of the rat vas deferens to field stimulation are adrenergic (Sjöstrand, 1981), then another interpretation of our results is that PGE_2 reduces noradrenaline release, whilst potentiating that action of noradrenaline on the tissue which gives rise to the slow component. This could explain the reduction of the fast component, with little effect on the slow component except for potentiation at high prostaglandin concentrations, where presumably the postjunctional effect outweighs the prejunctional effect. This interpreta-

tion would be consistent with effects of PGE_2 reported on the guinea-pig vas deferens, in which PGE_2 has been shown to reduce the stimulation-evoked noradrenaline overflow (Stjärne, 1973; Hedqvist, 1974) and also to potentiate the contractile effect of noradrenaline (Ambache & Zar, 1970; Hedqvist & von Euler, 1972; Petkov & Radomirov, 1980).

Blakeley, Brown, Cunnane, French, McGrath & Scott (1981) have shown recently that the fast, nifedipine-sensitive component of the response of the guinea-pig vas deferens to hypogastric nerve stimulation is associated with the generation of excitatory junction potentials (e.j.ps) which by facilitation and summation evoke muscle action potentials. This component of the response was resistant to α_1-adrenoceptor blockade. In contrast, the slow component of the response, which was sensitive to α_1-adrenoceptor antagonists, did not involve e.j.ps or propagated action potentials. If this is true also of the two components of the response of the rat vas deferens to field stimulation, then another interpretation of our results is that PGE_2 might interfere with the generation of e.j.ps or propagated action potentials, whilst having no effect on or (at high concentrations) potentiating the direct, α_1-mediated activation of the contractile process by noradrenaline.

We found PGE_2 to be considerably more potent in inhibiting the 'fast component' responses of the prostatic vas in Krebs solution containing 1.25 mmol/l Ca^{2+} (approximating to the normal plasma free ionized calcium concentration) than in the presence of a higher Ca^{2+} concentration (2.5 mmol/l). This was the case with preparations from both reserpine-treated and normal groups of animals, and is consistent with the influence of Ca^{2+} on PGE_2-induced inhibition of neurotransmission in other sympathetically innervated tissues (reviewed by Hedqvist, 1977). The potentiating effect of PGE_2 on the 'slow component' responses of the epididymal vas was also less marked at the higher Ca^{2+} concentration.

It was of interest that the preparations of prostatic vas from reserpine pretreated rats were considerably more sensitive to the inhibitory effect of PGE_2 than those from untreated rats; this was apparent particularly in the presence of the lower Ca^{2+} concentration (1.25 mmol/l Ca^{2+}). At present, there is no obvious explanation for this increased sensitivity of preparations from reserpine-treated rats to the inhibitory effect of PGE_2. As regards the experiments carried out at the lower Ca^{2+} concentration, it is possible that the greater effectiveness of PGE_2 on the preparations from reserpine-treated rats was simply a consequence of their lower contractility. However, this seems unlikely in view of the lack of correlation between the degree of PGE_2-induced inhibition and 'control' tension, in either reserpine-treated or normal preparations. Also, such an explanation would not be applicable to the results of the experiments carried out in 2.5 mmol/l Ca^{2+}, where the initial tensions developed by preparations from reserpine-treated and normal rats were very similar, but the prostaglandin still appeared to be more effective on the preparations from reserpine-treated animals.

The concentrations of PGE_2 used in our experiments are high in comparison with those reported to inhibit neurotransmission in the field stimulated guinea-pig vas deferens. However, this is in accordance with previous reports that vasa deferentia from rats are much less sensitive to prostaglandins than those from guinea-pigs (Ambache *et al.*, 1972; Hedqvist & von Euler, 1972).

In conclusion, the effects of PGE_2 on the two components of the response of the rat vas deferens to field stimulation are not as might be expected from its effects on sympathetic neurotransmission in several other tissues. Thus, the 'straightforward' adrenergic component of the response is not reduced, but rather is potentiated, by the prostaglandin, whereas the component that is either non-adrenergic, or at least does not show most of the properties associated with other adrenergic responses, is inhibited. The latter effect is the one typical of PGE_2 in tissues where sympathetically-mediated responses display the *usual* properties associated with adrenergic neurotransmission.

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