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THE RABBIT ANOCOCCYGEUS MUSCLE AND ITS RESPONSE TO FIELD STIMULATION AND TO SOME DRUGS

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SUMMARY

1. The response of the rabbit anococcygeus muscle to field stimulation of its intramural nerves and to some drugs has been examined and compared with results previously obtained in the rat and the cat.

2. The rabbit muscle possesses an adrenergic innervation as demonstrated histologically by the Falck and Hillarp fluorescence technique. This innervation is sparser than in the rat or cat.

3. *In vitro* the muscle usually shows little tone but if suitably stretched will develop a maintained contraction. The response to field stimulation depends on the level of tone. When this is low purely motor responses are obtained. In the presence of tone the response depends on its level and the frequency of stimulation; low frequencies are purely inhibitory, with increasing frequency the response becomes biphasic and high frequencies produce a purely motor response. The higher the tone the more prominent the inhibitory components. Guanethidine 10^{-5} M abolishes the motor component.

4. The muscle is caused to contract by noradrenaline, acting through α receptors, by histamine acting through H1 receptors and by 5-hydroxytryptamine. Part of the effect of the latter appears to be due to the release of noradrenaline.

5. The muscle is caused to relax by acetylcholine acting through muscarinic receptors, by isoprenaline through β receptors, by histamine in the presence of mepyramine through H2 receptors and by ATP and bradykinin.

6. The significance of these findings for the motor and inhibitory innervation of the muscle is discussed.

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INTRODUCTION

The properties of the anococcygeus muscle in the rat and cat have been described (Gillespie, 1972; Gillespie & McGrath, 1974*a*; Creed, Gillespie & Muir, 1975). Pharmacologically the muscle in the two species shows considerable differences but both have in common a motor adrenergic innervation and an inhibitory innervation the transmitter for which is as yet unknown. The rabbit possesses two well-developed smooth muscles related to the terminal colon, the caudo-sacral (anococcygeus) and the recto-coccygeus muscles (Langley & Anderson, 1896). The rectococcygeus possesses a motor cholinergic innervation but apparently no inhibitory nerves and responds to acetylcholine, noradrenaline and histamine by contraction (McKirdy, 1972). We have now examined the anococcygeus (caudo-sacral) muscle in the rabbit to see whether it possesses an inhibitory innervation comparable to the muscles in the other species and also to examine its response to drugs. Preliminary reports of some of these findings have been published (Creed & Gillespie, 1976).

METHODS

The anococcygeus muscles in the rabbit as in the rat are paired structures taking origin from the upper coccygeal vertebrae. The muscles lie behind the terminal colon but a cm or so before the anal margin pass on either side of the colon and appear to end within the longitudinal external muscle. Anatomically the arrangement differs in certain respects from the rat. The muscles are less discrete; at their caudal end they interdigitate with the centrally placed rectococcygeus as it leaves the distal colon and, finally, the two anococcygeus muscles do not unite in front of the colon as they do in the rat. This may indicate their lack of participation in the formation of a retractor penis muscle which in the rat takes origin from this ventral bar. This may also explain why the muscles are not heavier and stronger in male rabbits, a notable feature in male rats (Gibson & Gillespie, 1973).

Rabbits in the weight range 1·8–2·5 kg were killed by an overdoes of I.V. Nembutal. The anococcygeus muscles were identified as they lay behind the colon. The muscles were separated from one another and from the rectococcygeus muscle, each separately tied at its origin from the coccygeal vertebrae and just previous to its insertion into the colon, sectioned above and below these ties and removed to a 10 ml organ bath containing Krebs saline at 36 °C and gassed with a mixture of 95% O_2 and 5% CO_2. The muscles were initially stretched to a tension of 1 g and contractions recorded by a Grass FTO3 isometric transducer and displayed on a Grass Polygraph. In those experiments examining the response to field stimulation the muscles were threaded through a pair of ring platinum electrodes embedded in Araldite (Ciba). Stimulation was by 1 msec pulses at supramaximal voltage at the frequencies given in the text.

Drugs dissolved in saline were added to the bath in volumes from 0·1 to 0·5 ml., usually either 0·1 or 0·3 ml. The following drugs were used: acetylcholine chloride (Koch–Light), adenosinestriphosphate (ATP) (Sigma), atropine sulphate (Burroughs–Wellcome), guanethidine sulphate (Ciba), histamine acid sulphate (B.D.H.), 5-hydroxytryptamine creatinine sulphate (5-HT) (Koch–Light), isoprenaline hydro-

chloride (Sigma), mepyramine maleate (M. & B.), metiamide (SKF), methysergide bimaleate (Sandoz), noradrenaline bitartrate (NA) (Koch–Light), phentolamine mesylate (Ciba), propranolol (I.C.I.). Doses are given in moles of the base unless otherwise indicated.

The histological distribution of adrenergic nerves in the muscle was examined in freeze-dried blocks of tissue exposed to formaldehyde gas at 80 °C for 1 hr, embedded in wax, sectioned at 6 μm and mounted in liquid paraffin. The sections were examined in dark field conditions with exciting light from a mercury vapour lamp filtered through a 3 mm BG 12 exciter filter and with a 530 nm barrier filter above the microscope objective.

RESULTS

Adrenergic innervation

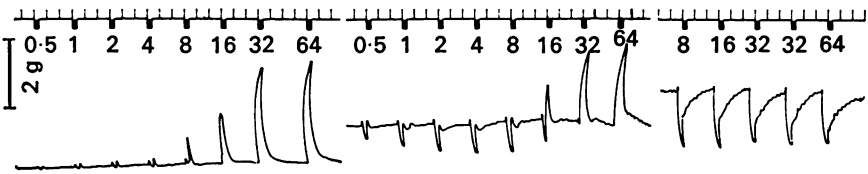
Pl. 1 shows the distribution and density of the adrenergic innervation of the rabbit anococcygeus as compared with the rat. The density of innervation is considerably less in the rabbit and the distribution on the outside of the muscle bundles suggests an organization of these into bundles of a cross sectional area greater than those in the rat. As in the rat the terminal innervation is uniform throughout the muscle length.

Spontaneous activity and the response to field stimulation

When stretched under the relatively mild tension of 1 g most muscles gradually relaxed over the next 20–30 min to a level of about 0·5–0·8 g and showed no rhythmic mechanical activity and drugs, such as acetylcholine or isoprenaline which inhibit the muscle, produced no apparent effect suggesting the residual tension was borne by passive elements in the tissue. A few muscles when stretched developed over the next 20 min an increase in tension and others developed rhythmic contractions. This activity was in response to stretch since, if this was slightly reduced, the rhythmic activity disappeared. If a greater degree of stretch was applied as described in the following article (Creed & Gillespie, 1977), then all preparations developed some degree of tone and rhythmic activity.

The response to field stimulation depended on the degree of tone. In the present experiments this was usually negligible so that only motor responses could be demonstrated. The frequency–response relationship was, therefore, relatively simple; all frequencies from single pulses upwards produced contraction and the maximum was not reached till about 64 Hz (Text-fig. 1). These responses are similar to those reported in the rat anococcygeus (Gillespie, 1972) other than the higher frequency needed to produce a maximum response and the occasional observation at the lower frequencies of a small inhibitory component to the response followed by a rebound contraction. The true nature of the effect of field stimulation was seen only when the muscle tone was raised to produce conditions appropriate for the display of either contraction or relaxation. It could

then be shown in every preparation that the response at low frequencies of stimulation (below 10 Hz) was primarily inhibition. As the frequency was increased the response changed from predominantly inhibitory to biphasic and, finally, to one mainly motor (Text-fig. 1). That the motor component was due to the stimulation of adrenergic motor nerves was shown by its disappearance on the addition of either phentolamine or guanethidine 10^{-6} M. Text-fig. 1 shows this effect for guanethidine.



Text-fig. 1. The response of the rabbit anococcygeus muscle to field stimulation at the frequencies in Hz shown below each event marker. In the first sequence the muscle possessed no intrinsic tone and the responses were predominantly motor with a maximum at 64 Hz. In the second sequence the muscle tone had been raised by the addition of histamine 10^{-6} M. The response to low frequencies of stimulation were now inhibitory, intermediate frequencies were biphasic and high frequencies motor. Guanethidine 2×10^{-5} M was then added; this blocked the motor adrenergic response and at the same time further raised tone, the response to stimulation at all frequencies was then inhibitory. Time marker 1 min.

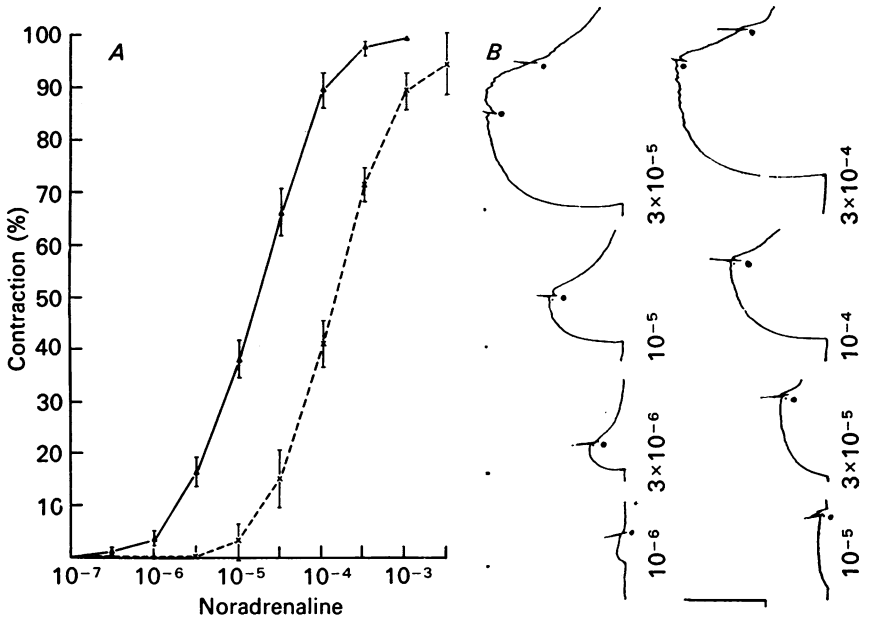
The response to drugs

Noradrenaline and isoprenaline

Noradrenaline caused a dose-related contraction of the rabbit anococcygeus muscle with a threshold at about 3×10^{-7} and a maximum at 3×10^{-4} M. The response consisted of a smooth, fairly rapid development of tone which, once fully developed, could be maintained for prolonged periods. Washing reversed the response but at high concentrations of the drug the decline in tension was slow in spite of repeated washing. Examples of these responses from one experiment are illustrated in Text-fig. 2. The response to NA was antagonized by phentolamine which produced a parallel shift in the dose–response curve with no change in the maximum which had a mean $\pm$ S.E. in the control of $5{\cdot}2 \pm 0{\cdot}3$ g (Text-fig. 2). In two experiments concentrations of NA, which before phentolamine had caused contraction, were found to produce an inhibitory response in its presence. Phentolamine also much reduced the prolonged contraction seen after washing out high concentrations of NA even when these concentrations were high enough to give maximal or near maximal responses.

Isoprenaline had no effect on the normal relaxed muscle until concentrations of 10^{-4} M were reached. This and higher concentrations caused

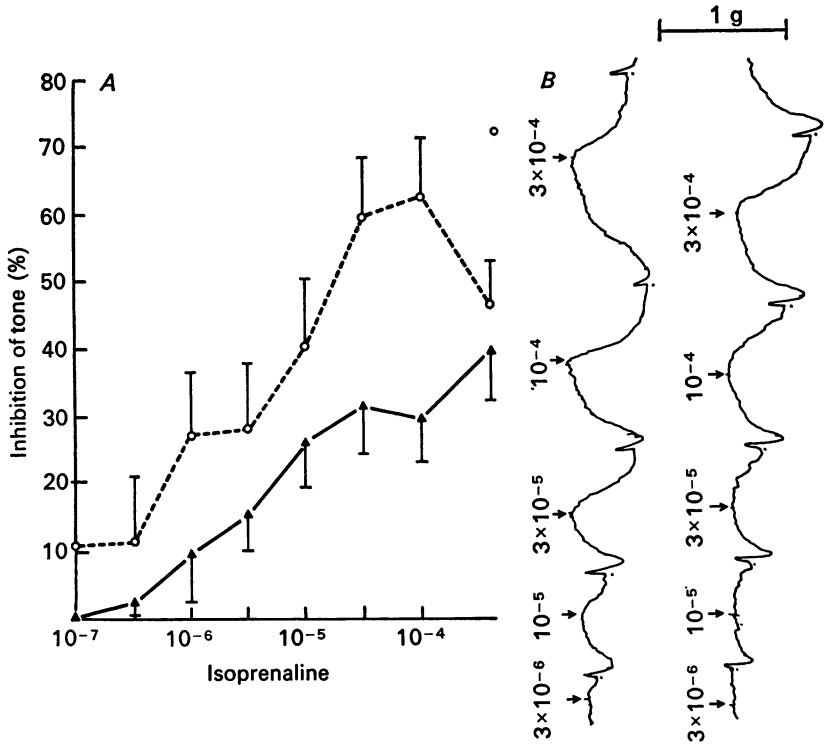
contraction, an effect abolished by phentolamine 2×10^{-6} M and unaffected by propranolol suggesting an action on α receptors. If muscle tone were raised either by guanethidine or histamine then isoprenaline caused a dose-related inhibition with a threshold of about 3×10^{-7} M. As the dose was increased the inhibitory response increased up to a concentration of about 10^{-4} M. At this and higher concentrations the inhibitory response either



Text-fig. 2. *A*, the dose–response curve to NA in the presence (× — ×) and absence (▲—▲) of phentolamine 2×10^{-6} M. Each point is the mean of six experiments and the bars represent ± 1 S.E. of the mean. Phentolamine displaces the dose response curve to the right by one order of magnitude without altering the maximum. *B*, the well-maintained nature of the response to NA. The upper row is control responses, the lower row responses in the same muscle in the presence of phentolamine 2×10^{-6} M. The black dots represent a wash. In the control the response at high concentrations of NA is not readily reversed by washing; in the presence of phentolamine, even where a contraction of comparable magnitude is produced by an appropriately increased concentration of NA, this effect is rapidly reversed by washing.

diminished or, on occasions, reversed to contraction. Once again this contractile component was abolished by phentolamine 2×10^{-6} M. In the presence of phentolamine the inhibitory response continued to increase up to a concentration of 3×10^{-4} M (Text-fig. 3*B*). The effect of propranolol 2×10^{-6} M on the inhibitory action of isoprenaline was studied in preparations in which tone was induced by histamine. The inhibitory response was

measured as the percentage inhibition of such tone as was present when the isoprenaline was added. Two groups were examined, one in which phentolamine 2×10^{-6} M was added in addition to histamine to abolish the α action of isoprenaline and the other was histamine alone. Analysis of these two groups showed no difference in their separate dose response curves except at concentrations of isoprenaline of 10^{-4} M or more, confirming our previous finding in atonic muscles that α stimulation requires high doses of isoprenaline. The two groups were, therefore, combined in constructing

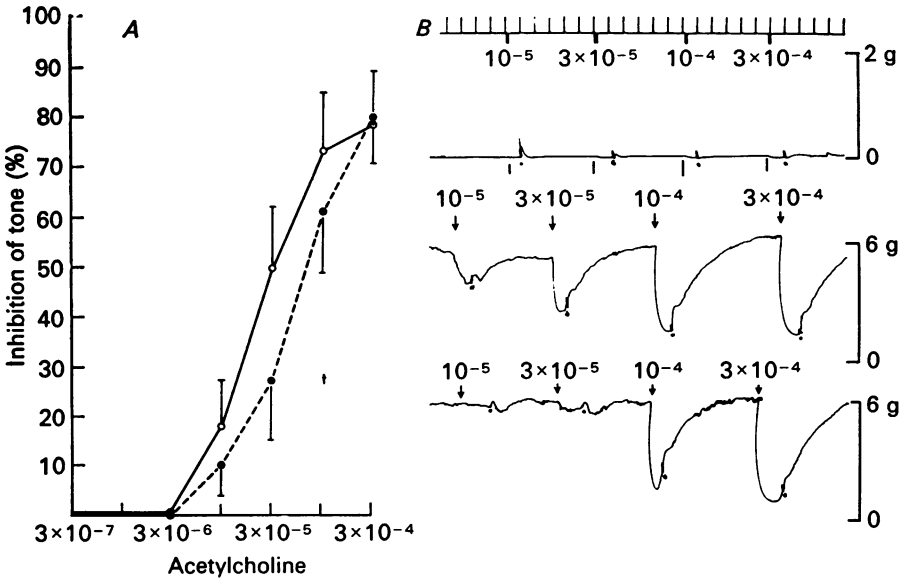


Text-fig. 3. *A*, the dose–response curves to the inhibitory effect of isoprenaline before (○ - - - ○) and in the presence (▲—▲) of propranolol 2×10^{-6} M. Tone was raised by adding histamine 2×10^{-6} M. Each value is the mean of between five and nineteen experiments and the bars represent ± 1 S.E. In most experiments phentolamine 2×10^{-6} M to block α receptors was present throughout. The dose–response curves are derived from experiments with or without phentolamine since the effect of isoprenaline on α receptors is only detectable at high concentrations of the drug. This effect on α receptors is responsible for the decline in inhibition at 3×10^{-4} M in the control. Propranolol displaces the dose response curve to the right. *B*, one example of the inhibitory responses to isoprenaline in a muscle whose tone was raised throughout by the presence of histamine 2×10^{-6} M. The upper traces are the control and the lower in the presence of 2×10^{-6} M propranolol.

the dose response curves shown in Text-fig. 3*A*. Propranolol 2×10^{-6} M inhibited the response to isoprenaline and shifted the dose–response curve to the right.

Acetylcholine

In preparations lacking tone, acetylcholine in concentrations from 10^{-7} to 3×10^{-4} M was without effect. If the tone was first raised with histamine then acetylcholine caused a dose-related inhibition with a threshold at



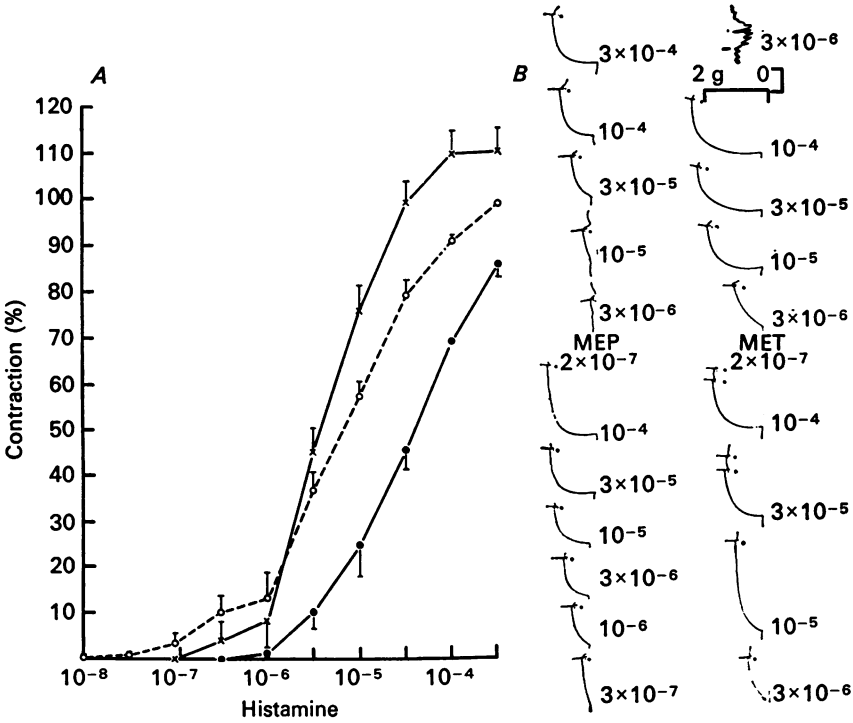
Text-fig. 4. *A*, dose–response curves to acetylcholine in the presence ● – – – ● and absence ○—○ of atropine 2×10^{-7} M. Each point is the mean of six observations and the bars represent the S.E. of these means. The dose response curve to acetylcholine is steep with a high threshold and is shifted to the right by atropine. *B*, one experiment to show that acetylcholine in an atonic muscle is without effect (upper trace) but when tone has been raised by histamine 2×10^{-6} M acetylcholine causes a rapid dose-related relaxation (middle trace) which is inhibited by atropine 2×10^{-7} M with no reduction in the maximum response (final trace). Time in minutes.

about 10^{-5} M and a maximum at about 3×10^{-4} M (Text-fig. 4). This dose–response curve in comparison with those for other agonists was steep and had a high threshold. Atropine 2×10^{-7} M shifted the dose–response curve to the right without altering the maximum response.

Histamine

In the rat and cat anococcygeus histamine is without effect. In the rabbit the drug proved an excellent agonist producing a dose-related con-

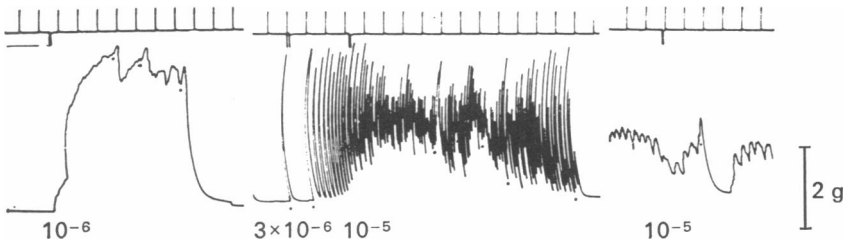
traction which was well maintained so long as the drug was left in contact with the tissue (Text-fig. 5). The threshold concentration was about 3×10^{-7} M with a maximum at 10^{-4} M. In spite of the higher sensitivity the maximum response of $2{\cdot}6 \pm 0{\cdot}6$ g was significantly less than that for NA. At high concentrations of histamine the response was slow to return to the base line on washing and this slow decay was not accelerated by repeated washing. The effect of the H1 receptor antagonist mepyramine and the



Text-fig. 5. *A*, dose–response curves to histamine in control muscles (○ - - - ○), in muscles in the presence of mepyramine 2×10^{-7} M (●—●) or in the presence of metiamide 2×10^{-7} M (× — ×). Each value is the mean of 12 observations in the control, and six observations in the presence of mepyramine or metiamide, and the bars are the S.E. Mepyramine shifts the dose response curve to the right, metiamide has no effect at low concentrations of histamine but moves the curve to the left at high concentrations and increases the maximum response. *B*, the responses to histamine in two different muscles and the effects of mepyramine 2×10^{-7} M (upper records) or metiamide 2×10^{-7} M (lower records). The responses to histamine are smooth well maintained contractions, mepyramine antagonizes the response without changing the maximum whereas metiamide increases the maximum response. The final response in the bottom row is from yet another muscle and shows the ability of histamine in spontaneously active muscle to cause inhibition. The horizontal bar represents 2 min.

H2 receptor metiamide on these responses was studied. As Text-fig. 5 shows, mepyramine antagonized the response shifting the dose response curve in a parallel fashion to the right. Mepyramine also abolished the prolonged response seen on washing out high doses of histamine in the same way that phentolamine abolished that to noradrenaline.

Metiamide 2×10^{-6} M had no ability to inhibit the response; indeed, in the presence of metiamide, there was a significant increase in the maximum response to $3{\cdot}8 \pm 0{\cdot}4$ g and the dose response curve was shifted to the left, an effect which increased with increasing concentrations of histamine. This result suggested the presence of H2 receptors occupation of which resulted in inhibition and, indeed, histamine in the absence of any blocking agent could occasionally, in muscles possessing high intrinsic tone, cause inhibition (Text-fig. 5).

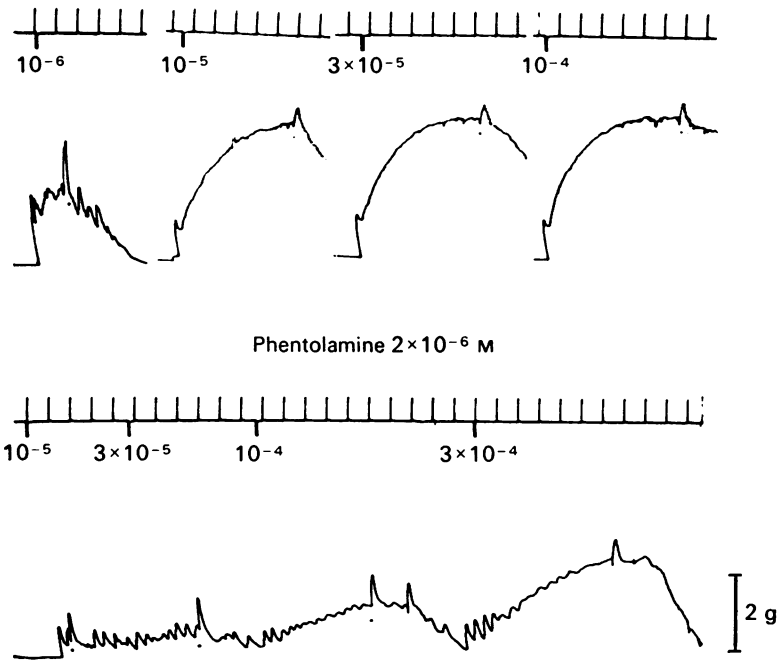


Text-fig. 6. Three variations in the response to 5HT in the molar concentrations shown beneath each record. The first trace illustrates the two component response which was most commonly observed, an initial slow contraction which appears to trigger a second and larger contraction, the second a muscle which responded by initiating rhythmic activity and the final record the rare observation of an inhibitory response. Time in minutes.

5-Hydroxytryptamine (5-HT)

This drug caused contraction of the muscle but of all drugs examined its effect was most variable both quantitatively and qualitatively. Quantitatively this appeared as variations both in the threshold for the onset of contraction, which ranged from 10^{-7} to 10^{-5} M, and the magnitude of the response. In different preparations the maximum contraction varied from 1·4 to 8·4 g with a mean and S.E. of $4{\cdot}1 \pm 0{\cdot}55$ g. Three qualitative variations in response were noted and are illustrated in Text-fig. 6. First, the responses to submaximal doses often showed two components, an initial slow contraction of relatively low amplitude which, after some delay, appeared to trigger a more rapid and larger contraction. Secondly, in some preparations 5-HT gave rise to an increase in rhythmic activity rather than a rise in tone and, thirdly, in a few preparations the response, especially to large doses, was inhibition. The possibility that the delayed rapid

component to the contractile response was due to the liberation of either NA from adrenergic nerves or histamine from mast cells was investigated by examining the effect of phentolamine 2×10^{-6} M and mepyramine 2×10^{-6} M on the response. Mepyramine had no effect but in the presence of phentolamine the response at all doses were reduced in amplitude and showed only one component (Text-fig. 7). It was not possible to be more

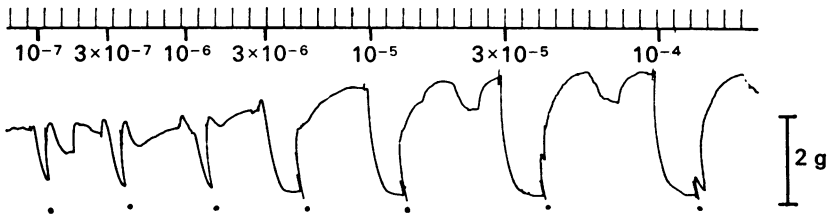


Text-fig. 7. The effect of phentolamine on the motor response to 5-HT. The upper records show the control responses to 5-HT in the molar concentrations shown beneath the event marker. Between the upper and lower records phentolamine 2×10^{-6} M was added to the bath. The response to 5-HT at all concentrations studied was reduced in the presence of phentolamine. Time in minutes.

precise in individual experiments since simple repetition of the same dose of 5-HT in the absence of phentolamine did not regularly produce responses of a similar configuration. Since phentolamine appeared to eliminate a possible adrenergic component, dose–response curves were constructed in the presence of phentolamine and the effect of adding methysergide 2×10^{-6} M examined. Methysergide displaced the dose–response curve to the right with little change in the maximum but this effect was small and not statistically significant. In those experiments in which 5-HT produced inhibition this effect was unaltered by methysergide.

Adenosine triphosphate (ATP)

In previous experiments ATP caused contraction in the rat but inhibition in the cat. In the rabbit the usual response was inhibition. In the absence of tone, however, the response was occasionally a dose-related contraction. In the presence of spontaneous tone or of tone induced by histamine, ATP was demonstrated to be a powerful inhibitory agent with a threshold about 10^{-7} M and a steep dose–response curve so that concentration of 10^{-5} M and above completely abolished tone. These effects are illustrated in Fig. 8.



Text-fig. 8. The response of the rabbit anococcygeus to increasing doses of ATP in the molar concentrations shown below the event marker. The tone of the muscle had been raised by the previous addition of histamine 2×10^{-6} M. The black dots indicate the removal of the drug. ATP exerts a powerful inhibitory effect at all concentrations. Time in minutes.

TABLE 1. The distribution of receptors and the responses they mediate in the rat, cat and rabbit anococcygeus muscle. The information on the rat and cat is from Gillespie (1972), Gillespie & McGrath (1974*a*), for prostaglandins from J. S. Gillespie & A. K. Tilmisany (unpublished) and for bradykinin (Gillespie & McKnight, 1976)

Drug	Receptor	Rat	Cat	Rabbit
Catecholamines	α	Motor	Motor	Motor
Catecholamines	β	0	Inhibitor	Inhibitor
ACh	Muscarinic	Motor	Inhibitor	Inhibitor
Histamine	H1	0	0	Motor
Histamine	H2	0	0	Inhibitor
5-HT		Motor	Motor	Motor
ATP		Weakly motor	Inhibitor	Inhibitor
Bradykinin		Inhibitor	Inhibitor	Inhibitor
Prostaglandins E_1 and E_2		Motor	Inhibitor	Not tested
Prostaglandin $F_{2\alpha}$		Motor	Inhibitor	Not tested

DISCUSSION

Comparison of the responses of the rabbit anococcygeus to drugs with previous results for the rat and cat emphasize the variability in the different species. This comparison is summarized in Table 1. The rat muscle responds to relatively few drugs and with the exception of bradykinin all

responses are motor. The cat and rabbit muscle have more in common, in particular the possession of inhibitory β adrenoceptors, inhibitory muscarinic receptors and an inhibitory response to ATP. These two species, however, are clearly distinguished by the presence in the rabbit of both H1 and H2 receptors for histamine, receptors which neither the cat nor the rat possess. These responses to drugs may throw some light on the possible nature of the transmitters of the motor and inhibitory nerves which exist in all three species. On the motor side all of the evidence is consistent with adrenergic transmission. The muscles in all three species contain nerve fibres which fluoresce with the technique of Falck and Hillarp, the content of NA is relatively high (Gillespie & McGrath, 1974*b*). All three species respond to NA by a maintained contraction and the response to nerve stimulation is abolished by both α blocking agents and guanethidine. The nature of the inhibitory transmitter by contrast is obscure. If it is assumed to be the same transmitter in all three species then several potential candidates such as acetylcholine and ATP can be ruled out because they are either ineffective or cause contraction in the rat muscle. The alternative, that in the rabbit and cat the transmitter is ATP but that another transmitter is responsible for inhibition in the rat though possible seems to us less likely. The only drug which inhibits in all three species, other than non-specific agents such as the nitrites and xanthine, is bradykinin. It is unlikely that this substance itself is the neurotransmitter but it may point to a peptide as a likely candidate.

The powerful inhibitory effect of ATP in the rabbit muscle raises the possibility that the inhibitory nerves may correspond to the intramural inhibitory nerves in the adjacent gut which have been postulated to produce their inhibition by the release of ATP (Burnstock, 1972). In the gut four features distinguished these non-adrenergic non-cholinergic nerves from the inhibitory extrinsic sympathetic nerves. First, their relative insensitivity to adrenergic neurone blocking drugs; secondly, the ability of a single pulse to cause mechanical inhibition; thirdly, nerve stimulation caused considerable hyperpolarization of the muscle membrane and, finally, following stimulation there was a rebound contraction (Burnstock, Campbell, Bennett & Holman, 1964). In previous experiments with the rat anococcygeus we failed to find any significant degree of hyperpolarization of the smooth muscle membrane even with repetitive stimulation at optimal frequencies and no evidence of rebound contraction (Creed *et al.* 1975). In the rabbit anococcygeus rebound contraction was an obvious feature (Text-fig. 1 illustrates this) and as the following article shows, mechanical inhibition is accompanied by membrane hyperpolarization and cessation of stimulation is followed by rebound contraction very like the responses observed in the gut. Nevertheless, it is difficult to see how ATP

can be the neurotransmitter in the anococcygeus unless one postulates a different transmitter in the rat since in this species ATP is motor.

The presence of histamine receptors in the rabbit muscle was unexpected since in other respects the drug sensitivity was identical to the cat which has no receptors for histamine. The finding was all the more surprising since the adjacent rabbit intestine is notably insensitive to histamine though interestingly neonatal rabbits up to 11 days old are highly sensitive to the motor effects of histamine and the receptors, as in the adult anococcygeus, are of the H1 type (Botting, 1975). Since the experimental demonstration of H1 and H2 receptors (Black, Duncan, Durant, Ganellin & Parsons, 1972) a great variety of tissues have been examined and it is clear that the existence of both types of receptor on the same effector cell is not uncommon. Both exist on blood vessels (Black, Owen & Parsons, 1975), on bronchial smooth muscle (Eyre, 1973), on ganglion cells (Brimble & Wallis, 1973) and on chick small intestine (Chand & Eyre, 1976). There is no consistent relationship between the receptor and the nature of the response as one finds for α and β adrenoceptors. In blood vessels in particular H1 and H2 receptors are inhibitory in the cat and dog (Black *et al.* 1975), H1 are constrictor and H2 dilator in the rabbit ear artery (Glover, Carroll & Latt, 1973), and in the coronary arteries of the guinea pig histamine apparently mediates a biphasic response through H1 receptors alone. Similarly, there is little consistency in the dominant response of the same tissue in different species. In tracheal smooth muscle, for example, histamine commonly causes contraction through an action on H1 receptors. In the sheep, however, the dominant response is relaxation mediated through H2 receptors and only in the presence of burimamide is the existence of bronchoconstrictor H1 receptors revealed (Eyre, 1973). The situation in the rabbit anococcygeus is the reverse of this, dominant contraction-mediating H1 receptors and inhibitory H2 receptors uncovered only in the presence of mepyramine. The functional significance of these receptors is obscure.

In its response to field stimulation the rabbit muscle differs in some respects from both rat and cat muscle. First, motor responses are less prominent in the rabbit, probably a reflexion of the lower density of adrenergic nerves and the greater degree of spontaneous activity which serves as a background for the display of inhibition. Secondly, after-contractions following inhibitory responses were a prominent feature but were never seen in the muscles in other species and, thirdly, the optimum frequency for motor responses was high, 64 Hz. The reasons for this difference in frequency sensitivity are unclear. It may be related to the different balance between inhibitory and motor innervation or to the longer diffusional pathway in the rabbit.

The properties of the anococcygeus also differ strikingly from the closely adjacent rectococcygeus muscle which has no inhibitory innervation, in which ATP as well as ACh mediate contraction and in which the motor innervation is cholinergic (McKirdy, 1972). It may be that in the defaecation reflex these muscles have a reciprocal action and that the anococcygeus is required to relax while the rectococcygeus shares in the contraction of the longitudinal muscle of the colon and by its attachment to the coccygeal vertebrae finally transfers that tension to bone.

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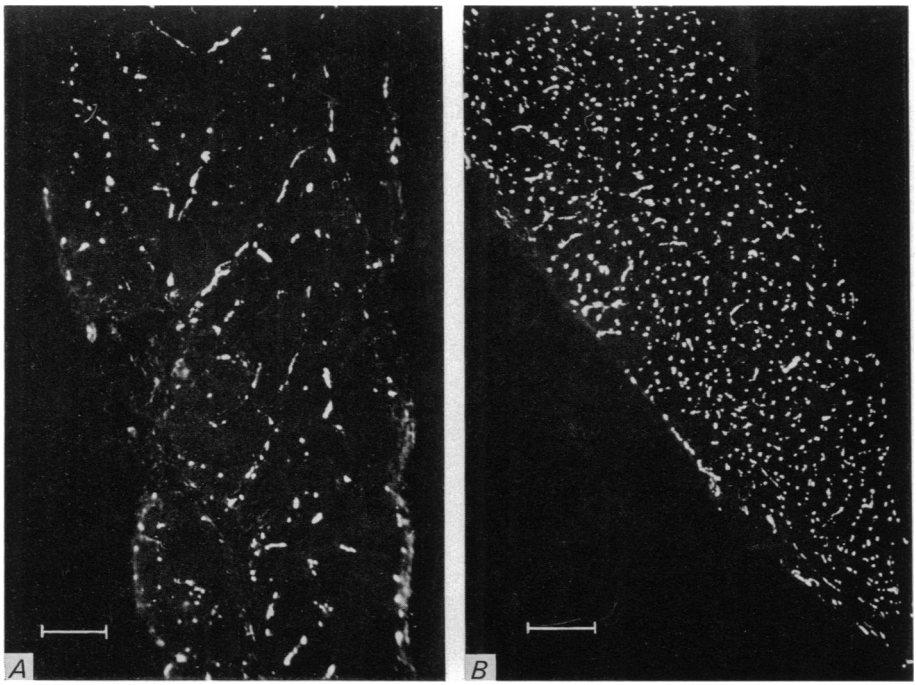
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EXPLANATION OF PLATE

Fluorescence in transverse sections of rabbit (*A*) and rat (*B*) anococcygeus muscles after treatment by the Falck and Hillarp technique. In both muscles the brightly fluorescent terminal adrenergic nerve fibres are uniformly distributed but the density of innervation is much greater in the rat. The individual nerve bundles are somewhat larger and the non-fluorescent muscle bundles they outline are considerably larger in the rabbit. The bar in each photograph represents 150 μm.