Species variation in motor transmission to the retractor penis muscle

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The contractions elicited by transmural stimulation (1-16 pulses; 0.2 ms; 10 Hz) of isolated retractor penis (RP) strips from dogs, pigs, sheep and horses were abolished by tetrodotoxin (1.6 μM) and were therefore neurogenic.

Although, in all species, motor responses to noradrenaline (0.6-3 μM) or tyramine (12-58 μM) were abolished by phentolamine (5.3 μM) or phenoxybenzamine (2.9 μM), the susceptibility of the neurogenic contractions to α-blockade was subject to species variation. In dog RP, the whole of the motor response was promptly abolished by phentolamine or phenoxybenzamine. But in pig RP phentolamine never abolished and only slightly reduced these responses (by $< 5\%$ with single pulses and $< 34\%$ with 16 pulses), without further reduction on raising the phentolamine concentration to 26.5 μM or on switching to phenoxybenzamine (2.9 μM) for 1 h (Figure 1); or on addition of propranolol (3.4 μM), atropine (2.9 μM), mepyramine (1.25 μM) or dimethyltubocurarine (23 μM).

In pig RP the phentolamine-resistant sympathetic motor transmission was reduced but never completely abolished by bretylium (48 μM) or guanethidine (40 μM); there was a tetrodotoxin-susceptible 10% remnant even after 80 μM guanethidine. Reserpine pretreatment (10 mg kg^{-1} day^{-1} for 3 days) abolished responses to tyramine (105 μM) but not the neurogenic contractions (4-16 pulses) which were wholly phentolamine-resistant.

A considerable phentolamine-resistant component was also found in sheep but not in horse RP.

Thus, whereas in the dog and horse the motor transmission to the retractor penis is wholly adrenergic, as found by previous workers, in the pig and sheep a large proportion of the motor response is unaffected by α-adrenoceptor blockade and appears to resemble the non-adrenergic type of motor transmission found in the vas deferens of many species (Ambache & Zar, 1971; Ambache, Dunk, Verney & Zar, 1972; Euler & Hedqvist, 1975; Jenkins, Marshall & Nasmyth, 1975).

In phenoxybenzamine-treated pig RP, twitch-inhibition by noradrenaline (6 μM) was blocked by propranolol (3.4 μM).

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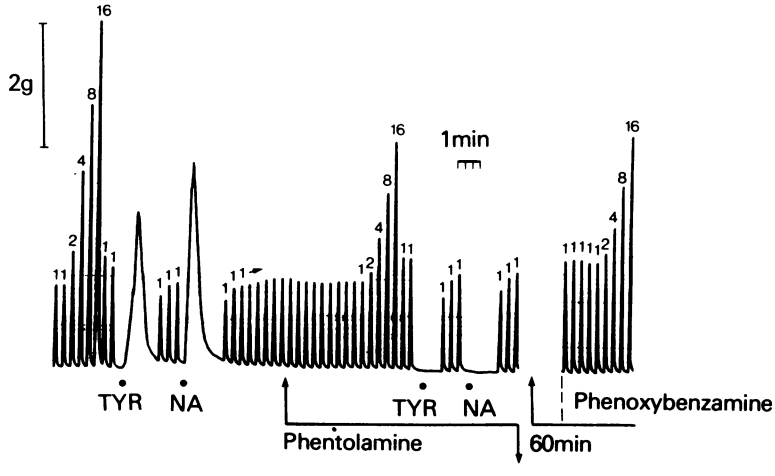

Figure 1 Pig RP. Persistence of a large proportion of the motor responses to 1-16 pulses (0.2 ms; 10 Hz; 1 min intervals; train length indicated by superscripts) in phentolamine (26.5 μM) and then in phenoxybenzamine (2.9 μM for 1 h). At the dots, tyramine (TYR; 26 μM) or noradrenaline (NA; 1.5 μM) for 60 seconds.

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Effects of morphine on acetylcholine release from the frog spinal cord

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Varying reports on the effects of morphine on acetylcholine (ACh) release from the cerebral cortex have been made. For example, in the cat morphine either depresses (Jhamandas, Phillis & Pinsky, 1971) or stimulates (Phillis, Mullin & Pinsky, 1973) ACh release. The opiate action on cortical cholinergic fibres is probably indirect but further experiments are needed to clarify this matter. To this end the frog spinal cord was chosen as an *in vitro* CNS preparation in order to study the effects of bath-applied morphine on spinal root potentials and endogenous ACh release with the method previously reported (Nistri, 1975). The effects of morphine on the ACh content of the frog brain and spinal cord *in vivo* have already been reported (Nistri, Pepeu, Cammelli, Spina & De Bellis, 1974).

The spontaneous ACh release from the frog cords was 7.3 ± 1.06 ng/ml every 10 min (mean ± s.e. mean, $n = 28$). The effects of morphine were variable and related to the concentrations used. Ten minutes after the application of 1 μM morphine spontaneous ACh release was reduced by $29.7 \pm 5.5\%$; a similar decrease was also found in subsequent samples. However, with this dose of morphine a 10 min antidromic ventral root stimulation (1 Hz; 0.1 msec; supramaximal voltage) was accompanied by a $246.8 \pm 70.1\%$ increase in ACh output over preceding values whereas in untreated cords similar stimulation yielded a $177.7 \pm 50.7\%$ rise. The dorsal root potential produced by such stimulation was slightly reduced by 1 μM morphine. All these effects were reversible on washing. Ten minutes after 100 μM morphine was added to the bathing fluid a $142.2 \pm 43.2\%$ rise in unstimulated ACh output was seen; the increase persisted in the following samples. However ventral root stimulation could not produce any further rise in ACh output. A small increase in ventral as well as dorsal root potentials was found. Naloxone (100 μM) slightly stimulated the spontaneous ACh release ($+ 51.4 \pm 15.3\%$), prevented the decrease in ACh output following 1 μM morphine and reduced the stimulation of ACh release after high doses of morphine.

In the frog spinal cord the motor axon collaterals are cholinergic fibres (Mitchell & Phillis, 1962) which can be directly activated by antidromic ventral root stimulation. Since small concentrations of morphine depressed spontaneous ACh output but failed to reduce the electrically-evoked ACh output, it is suggested that morphine reduced ACh release through an indirect mechanism probably mediated by interneurones. The stimulant action of high concentrations of morphine on spinal ACh release and root potentials might be one of the factors involved in the behavioural excitation seen in frogs after large doses of this opiate.

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