Short communications

Adrenoceptors in the human foetal small intestine

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Human foetal small intestine was shown to contain both α- and β-adrenoceptors with a predominance of β-adrenoceptors. The tissue examined was obtained from foetuses of gestational age between 8 and 26 weeks.

Both α- and β-adrenoceptors are present in the small intestine of the adult human (Bennett, 1965; Coupar & Turner, 1969; Hedges & Turner, 1969) and stimulation of either receptor produces a relaxation. McMurphy & Boreus (1968) concluded that only β-adrenoceptors are present in human foetal ileum. We reported (Hart & Mir, 1970) that phenylephrine relaxes human foetal intestine, and evidence is presented here that two types of adrenoceptors are present in the foetal tissue.

Methods.—The whole of the foetal intestinal tract was obtained from the Tissue Bank, Royal Marsden Hospital. The tissue was removed from the foetus by the staff of the Tissue Bank 2–4 h after hysterotomy had been performed for the legal termination of pregnancy. The tissue was stored in Krebs solution at 4° C. Two–seventeen hours after removal from the foetus 2–3 cm of small intestine was suspended in a 10 ml bath containing Krebs solution bubbled with 5% CO_2 in O_2 at 37° C and the tone and movements of the longitudinal muscle were recorded with an isometric transducer under a tension of 1–2 g. Most preparations were of ileum but in some of the preliminary experiments the jejunum was used. The tissue was allowed to equilibrate for 2 h before the administration of drugs.

Reproduceable relaxations could be obtained with tissues from the more mature foetuses (18 weeks) when the sympathomimetic amines were given at 6 min intervals, but with the younger tissue (11 weeks) it was necessary to use a time cycle of 15–20 minutes.

A contact time of 5 min was allowed for phentolamine, and 10 min for propranolol, before the administration of the test amine.

The drugs used were acetylcholine chloride, noradrenaline bitartrate, isoprenaline hydrochloride, phenylephrine hydrochloride, phentolamine methylsulphate, and propranolol hydrochloride. Drug concentrations are expressed as the final bath concentration of free base in g/ml.

Results.—Most preparations of small intestine showed spontaneous activity throughout the experiment and all contracted in the presence of acetylcholine (10^{-6} to 10^{-8}M). Each of the sympathomimetic amines inhibited spontaneous activity and produced a relaxation of the longitudinal muscle.

Responses to noradrenaline were studied on tissue from over fifty foetuses of gestational age between 8 and 26 weeks. All the preparations responded to concentrations of 5×10^{-9} to 5×10^{-7}M and preparations from the older foetuses tended to be more sensitive. Propranolol, in concentrations of $1{\cdot}8 \times 10^{-7}$ to $1{\cdot}8 \times 10^{-6}$M, inhibited the response to noradrenaline on twelve preparations. The effect of phentolamine (4×10^{-7} to 2×10^{-6}M) on the response to noradrenaline was investigated on twelve preparations: on eight the α-adrenoceptor blocking agent was without effect whilst on four the relaxation was reduced by up to 60%.

On twelve preparations from foetuses of gestational age between 11 and 20 weeks, phenylephrine was effective in concentrations of 5×10^{-8} to 4×10^{-7}M. On two a small contraction was obtained. The relaxation due to phenylephrine was blocked only partially by a concentration of propranolol which completely inhibited the response to noradrenaline. Phentolamine (3×10^{-7} to 8×10^{-7}M) inhibited the phenylephrine response (nine preparations) and thus had a greater effect on phenylephrine responses than on noradrenaline responses. On some preparations phenylephrine caused a small contraction in the presence of phentolamine (Fig. 1).

Responses to isoprenaline ($1{\cdot}3 \times 10^{-9}$ to 2×10^{-7}M) were obtained on tissues from seventeen foetuses of gestational age between 11 and 25 weeks. The response to isoprenaline was inhibited by propranolol (2×10^{-7}M) on four preparations but a concentration of phentolamine (3×10^{-7}M) which blocked the response to

phenylephrine produced little inhibition of the isoprenaline relaxation on six.

In a few experiments the amine was added while the tone of the preparation was raised with acetylcholine; this procedure did not alter the relaxation response to the amines or the activity of phentolamine or propranolol.

The responses of the ileum from a 20-week foetus are shown in Fig. 1. Propranolol (10^{-6}M) inhibited the response to isoprenaline by about 80%, that to noradrenaline by about 50% but had no effect on the phenylephrine response. In contrast, phentolamine ($7{\cdot}5 \times 10^{-7}$M) reduced the responses to isoprenaline and noradrenaline by only about 20% but blocked completely the relaxation to phenylephrine. The actions of phentolamine were studied on twenty-one preparations and eight of these contracted on addition of the phentolamine.

In the absence of either inhibitor the order of potency of the three amines was isoprenaline>noradrenaline>phenylephrine.

Discussion.—Both the order of potency of the three sympathomimetic amines and the selective inhibition produced by propranolol and phentolamine show that the human foetal small intestine contains β-adrenoceptors. In addition, the sensitivity of the phenylephrine relaxation to inhibition by phentolamine, and its resistance to propranolol, is good evidence that α-adrenoceptors are also present in the foetal small intestine. This is contrary to the conclusion of McMurphy & Boreus (1968) who reported that only β-adrenoceptors are present in human foetal ileum. Their conclusion was based on an order of potency in which isoprenaline>adrenaline=noradrenaline$\gg$methoxamine, plus the finding that, apart from methoxamine, the responses to the amines were inhibited by propranolol but not by phenoxybenzamine.

The relaxations produced by methoxamine were unaffected by either α- or β-adrenoceptor blocking agents. The gestational age of the tissues examined by McMurphy & Boreus was similar to that in the present study but their experimental procedure differed in two respects: they used a Tyrode solution at 37° C which had a pH of 7·4, and relaxations were obtained during an acetylcholine induced contraction. In the present study Krebs solution, with a pH of 7·4 was used at 37° C. The possibility that the small differences between the composition of the two solutions might explain the different results has not been investigated but seems unlikely. We found that phenylephrine was capable of relaxing the preparation which had contracted in response to acetylcholine and that this relaxation was sensitive to phentolamine. It appears, therefore, that the foetal intestine contains adrenoceptors which are sensitive to phenylephrine and phentolamine but not to methoxamine.

Read & Burnstock (1970) used fluorescence microscopy to study the adrenergic innervation of human foetal intestine of gestational age between 5 and 19 weeks. Fluorescent nerves first appeared in Auerbach's plexus at 9–10 weeks and, in smaller numbers, in Meissner's plexus at 13 weeks. Incubation studies showed that the intestine from an 8-week foetus could take up noradrenaline although it did not contain sufficient endogenous noradrenaline for fluorescence. Tissue from the one 8-week foetus examined in the present study gave dose dependent relaxations to noradrenaline suggesting, at least, that

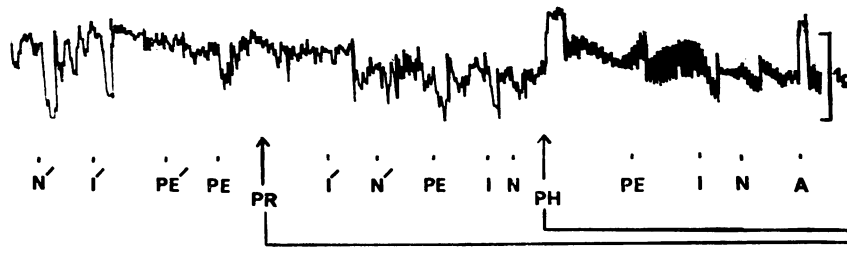

FIG. 1. Small intestine from a 20 week foetus. Responses to noradrenaline 5×10^{-9}M (N'), 10^{-8}M (N); isoprenaline, $1{\cdot}3 \times 10^{-9}$M (I), $8{\cdot}5 \times 10^{-9}$M (I); phenylephrine, 2×10^{-7}M (PE), 4×10^{-7}M (PE); acetylcholine, 4×10^{-8}M (A); propranolol, 10^{-6}M (PR) and phentolamine, $7{\cdot}5 \times 10^{-7}$M (PH).

some adrenoceptors are present by the time the first adrenergc fibres have developed.

In the foetal small intestine β-adrenoceptors predominate, as reported by McMurphy & Boreus and confirmed in our study, and in this respect the foetal tissue differs from that of the adult which contains both α- and β-adrenoceptors (Bennett, 1965; Coupar & Turner, 1969; Hedges & Turner, 1969). This developmental difference indicates that the α- and β-adrenoceptors are distinct entities and it is tempting to suggest that they are located on separate structures within the intestinal wall. This concept was first proposed by Kosterlitz and Watt (1965) who suggested that in the guinea-pig ileum the α-adrenoceptors are associated with nerve plexuses whilst the β-adrenoceptors are located on the smooth muscle. Although there is further experimental support for this concept the evidence does not allow a definitive conclusion to be drawn (Lee, 1970). If, however, this distinction is applicable to human tissue it is not unreasonable to suggest that the β-adrenoceptors associated with the muscle should be functional at an earlier stage of foetal development than the α-adrenoceptors whose activity is dependent upon the development of the nerve plexus.

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