An isolated perfused coronary artery preparation from the kitten

Previous unpublished observations from this laboratory indicated that coronary vessels in the kitten heart perfused by Langendorff's method develop more spontaneous tone than those in perfused hearts from cats (>1·5 kg), rabbits or guinea-pigs. This led us to examine the isolated coronary arteries from the kitten for the study of coronary vasodilators.

Kittens of either sex, 500–850 g, were anaesthetized with ether and the heart removed and placed in cold McEwen solution (1956) gassed with 5% carbon dioxide in oxygen. The aorta was quickly cannulated and the heart perfused by Langendorff's method. The anterior descending branch of the left coronary artery, being superficially located, was then readily dissected free from the myocardium and all side branches ligated. The vessel was cannulated with a 23 gauge needle from which the tip had been removed. A 1 to 1·5 cm piece of the artery was cut away from the heart and perfused as described for the rabbit ear artery preparation (De La Lande & Rand, 1965) with McEwen solution maintained at 32° and gassed with 5% carbon dioxide in oxygen. The initial perfusion rate was 5 ml min^{-1} and this was gradually increased stepwise in 0·5 ml min^{-1} increments over 30 to 60 min until the artery began to develop tone. The maximum perfusion rates used were 7 to 8 ml min^{-1}. When the tone became constant single doses of the compounds under test were injected in a volume of 0·05 ml into the rubber tube connecting the arterial cannula to the perfusion apparatus.

Most preparations developed a resistance to perfusion of 30 to 90 mm Hg. This is much higher than the values of 3 to 20 mm Hg reported for isolated coronary arteries of the dog perfused with McEwen solution (Trinker, 1973). It was therefore possible to study coronary dilators without using spasmogens as have been employed in most other recent studies of isolated coronary artery preparations (see for example Bohr, 1967; Baron, Speden & Bohr, 1972; Norton, Gellai & Detar, 1972; Johansson, 1973).

Dilatation of the isolated coronary artery was produced by acetylcholine (0·7 to 4×10^{-10} mol), nicotine (2 to 4×10^{-9} mol), histamine (1 to $1{\cdot}5 \times 10^{-9}$ mol), bradykinin (0·5 to 2×10^{-10} mol), adenosine (0·5 to 4×10^{-9} mol) and adenosine triphosphate (0·2 to $1{\cdot}8 \times 10^{-9}$ mol). Threshold dilator doses given above were found in 4 or more preparations. These took some time to establish for nicotine, bradykinin and the adenine nucleotides due to increases in the sensitivity of the artery to repeated injections of these compounds. Dilatation of the artery was also produced by β-adrenoceptor agonists. Comparisons of threshold doses showed that the artery was more sensitive to isoprenaline (0·5 to 1×10^{-11} mol) than noradrenaline (0·5 to 1×10^{-10} mol) or adrenaline (1·5 to 3×10^{-10} mol). A section of a trace in which several doses of isoprenaline and adrenaline were tested for

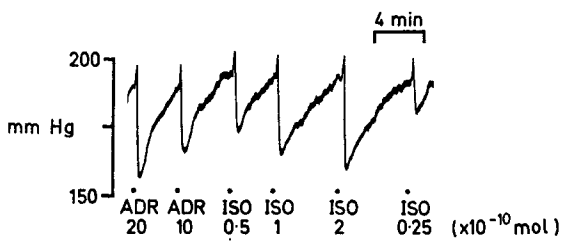

FIG. 1. Responses of a kitten isolated coronary artery preparation to adrenaline (ADR) and isoprenaline (ISO). Doses are expressed as 10^{-10} mol.

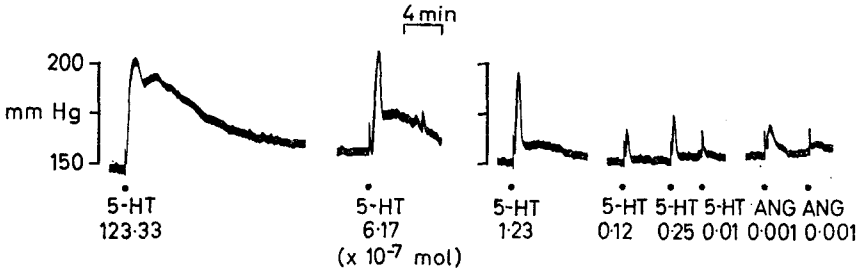

FIG. 2. Traces from an experiment in which the effects of 5-hydroxytryptamine (5-HT) and angiotensin (ANG) were tested on a kitten isolated coronary artery preparation. Doses are expressed as 10^{-7} mol.

their dilator effects is shown in Fig. 1. When large doses of the amines were tested in the presence of propranolol (1×10^{-6}M) at a concentration which antagonized the dilator responses of the amines, no α-adrenoceptor mediated constrictor responses were obtained. Similarly no constrictor responses could be obtained with phenylephrine (1×10^{-6} to 1×10^{-5} mol) or oxymetazoline (1×10^{-6} to 1×10^{-5} mol) which are more specific α-adrenoceptor agonists (Mujic & van Rossum, 1965). Both compounds produced coronary vasodilatation in doses greater than 1×10^{-6} mol. A vasodilator response to oxymetazoline may occur in other arterial beds (Hotovy, Enenkel & others, 1961).

The anterior descending branch of the left coronary artery in the kitten thus appears to contain β-adrenoceptors but few, if any, α-adrenoceptors. It therefore differs from the corresponding arteries of man (Andersson, Holmberg & others, 1972) and dog (Mekata & Niu, 1969; Trinker, 1973) which contain both α- and β-adrenoceptors and in that dilator responses are obtained with acetylcholine and histamine which constrict the coronary arteries of other species (Smith, 1950; Carrier, 1965; Norton & others, 1972). As in the kitten, adenosine also had a dilator effect on preparations from the rabbit (Norton & others, 1972) and dog (Walter & Bassenge, 1968). 5-Hydroxytryptamine (5-HT) and angiotensin (Fig. 2) produced an initial constrictor response on the kitten preparation but there was a marked tachyphylaxis to angiotensin (Fig. 2) whilst in some preparations repeated administration of 5-HT led to the production of dilator responses.

Both substances produced constrictor responses in dog coronary arteries (Bohr & Johansson, 1966).

With the exceptions of angiotensin and 5-hydroxytryptamine, the substances tested gave reproducible responses. This, together with the ease of dissection and maintenance of spontaneously developed tone for at least 4 h, make the kitten isolated coronary artery a good preparation for studying coronary vascular reactivity to drugs. In particular, the paucity of α-adrenoceptors in the artery should make it a suitable tissue for comparing coronary vasodilator potencies of β-adrenoceptor agonists.

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March 8, 1974

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The conformation of dopamine at its uptake site; further studies with rigid analogues

Recently various attempts have been made to obtain information about the conformation at the uptake site of inhibitors of the neuronal catecholamine transport system by the use of less flexible analogues (Horn & Snyder, 1972; Miller, Fowble & Patil, 1973; Tuomisto, Tuomisto & Swissman, 1974). In the case of dopamine the three most important conformations to be considered are the *trans* or *anti* form, Fig. 1a, and the two *gauche* forms, Fig. 1b and c. Through the use of *cis*- and *trans*-2-phenylcyclopropylamine and 1- and 2-aminoindane it was suggested that in homogenates of the rat brain corpus striatum and hypothalamus the preferred conformation for a non-catechol inhibitor of dopamine or noradrenaline uptake, such as amphetamine, was *trans* rather than *gauche* (Horn & Snyder, 1972). This conclusion was supported by studies on the inhibition of noradrenaline uptake in the rat vas deferens by *cis*- and *trans*-3-phenyl-2-methylazetidin-3-ol and *cis*- and *trans*-2-phenylcyclopropylamine (Miller & others, 1973). Tuomisto & others (1974) have prepared and tested various *trans*-decalin derivatives which contain the catecholamine moiety and have published evidence that the preferred conformation for the interaction of catecholamines with the uptake site in rat brain is *gauche*. In order to obtain further information on this topic two other rigid analogues of dopamine, 2-amino-6,7-dihydroxy-1,2,3,4-tetrahydronaphthalene (ADTN), Fig. 1d, and 6,7-dihydroxytetrahydroisoquinoline (norsalsolinol), Fig. 1e, have been examined as inhibitors of [^{3}H]dopamine (^{3}H-DA) uptake in synaptosome rich homogenates of the rat corpus striatum.

Adult male Sprague-Dawley rats were pretreated 18 h previously with reserpine (5 mg kg^{-1}) to inactivate the granular catecholamine storage mechanism. The effect

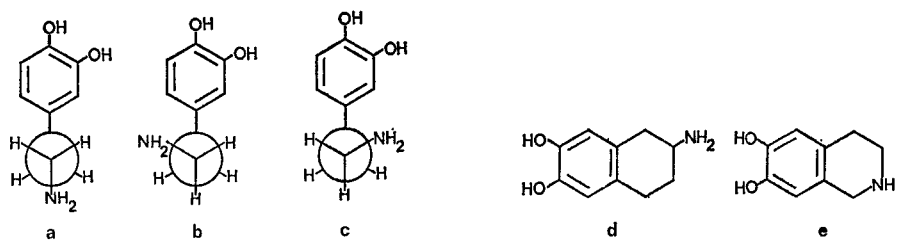

FIG. 1. Newman projections of the *trans* conformation of dopamine (a) and the two gauche conformations (b and c). Structural formulae for 2-amino-6,7-dihydroxy-1,2,3,4-tetrahydronaphthalene (ADTN) (d) and 6,7-dihydroxytetrahydroisoquinoline (e).