In our studies male C.S.E. rats 170–270 g were used ; analysis was made of total conjugated adrenaline and noradrenaline, using the methods of Anton & Sayre (1962) and Moelottke & Sloan (1970).

Total normetadrenaline and metadrenaline (Anton & Sayre, 1966) and H.M.P.G. and hydroxymethoxymandelic acid (Ceasar, Ruthven & Sandler, 1969) was also carried out including standards for each substance, on two 24 h samples of urine, collected using glass metabolism cages (26 cm diameter Metabowl).

Differences in the urinary excretion of catecholamines will be demonstrated in groups of rats maintained under different experimental conditions.

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Estimation of angiotensin II blood concentrations in hypertensive rats

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Many factors are thought to be associated with the development and maintenance of experimental hypertension. Previous work in these laboratories (Finch & Leach, 1970) has been concerned with the role of the sympathetic nervous system in renal and metacorticoid hypertension in the rat, and present studies are concerned with the role of vasoactive substances in experimental hypertension. In particular, the levels of circulating angiotensin II are being investigated using the superfused isolated organ technique (Vane, 1964 ; Regoli & Vane, 1964). Angiotensin II estimation using the isolated ascending colon of the rat, is made with the preparation bathed with either blood or Krebs solution. In the case of blood superfused organs, modifications have been made to the original procedure to deal with the small blood volumes available in the rat.

Fluid is pumped by a Watson-Marlow flow-inducer, at a rate of 2 ml/min via polythene tubing (Portex Ltd.) of internal diameter 0·584 mm $\pm$ 0·076 mm ; total extracorporeal volume of the tubing, including that in the pump, is not greater than 1 ml. The whole system is cleaned and sterilized by the method described by Vane (1971).

Angiotensin II in large rats (>200 g) is assayed by superfusing the tissue directly with the rat's blood. The assay tissue is washed with Krebs solution prior to switching to blood superfusion through a three way tap.

Calibration of the tissue responses is achieved by infusion of angiotensin II solution into the superfusing blood obtained from an acutely, bilaterally nephrectomized anaesthetized rat, and therefore, free of naturally occurring angiotensin II.

Interference from catecholamines is eliminated by the use of a suitable β-adrenoceptor blocking drug, infused intraluminally on to the isolated colon. Levels are also monitored using the isolated chick rectum.

Blood samples from rats weighing less than 200 g are taken from the abdominal aorta under ether anaesthesia ; the sample is centrifuged at 1,600 g for 8 min and the plasma (0·1 ml aliquots) injected into superfusing Krebs solution. In this case, the tissue responses are calibrated by injection of 0·1 ml aliquots of angiotensin II dissolved in plasma from bilaterally nephrectomized rats.

This technique allows the routine estimation of angiotensin II-like activity in experimental and spontaneous hypertension at all stages of its development.

This work was supported by the M.R.C.

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The isolated blood perfused rat tail preparation

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The isolated rat tail preparation provides a convenient and easily accessible ganglion-free vascular bed for the study of pharmacologically active compounds and the vascular responses to sympathetic nerve stimulation (Wade & Beilin, 1970).

The modification which is to be demonstrated, involves the perfusion of the tail vascular bed with blood drawn from, and returned to, a donor rat.

Drugs may thus be administered either directly via the arterial cannula into the vascular bed of the isolated tail, or may be administered to the donor rat at dose levels equivalent to those used for effecting cardiovascular activity in the intact circulation. In the latter case it would be expected that the agents so administered would be delivered, in due course, to the isolated rat tail vascular bed at pharmacologically active blood concentrations.

This technique thus enables combined *in vitro* and *in vivo* examination of drug activity. The demonstration will show the effects of four anticholinesterase compounds administered intravenously to the donor rat, at their commonly employed dose levels, on the responses to sympathetic periarterial nerve stimulation.

It will be seen that eserine (0·4 mg/kg) and BW284C51 (0·4 mg/kg) markedly enhanced the responses to nerve stimulation (3–12 Hz) whilst neostigmine 0·2 mg/kg) and Ro 02–0683 (0·2 mg/kg) have little effect.

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