DEMONSTRATIONS

A method for studying baroreflex bradycardia in the unanaesthetized rat

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The 'bolus method' of Smyth , Sleight & Pickering (1969) for quantifying baroreflex bradycardia in man is unsuitable for use in the rat, mainly because of a latency of response to the rapid blood pressure elevation (Redfern & Little, unpublished; Coleman, 1980). We have developed an infusion method which overcomes this problem.

Male Wistar rats (240–290 g) were prepared under ether anaesthesia (approx. 25 min). Mean arterial pressure (MAP) was recorded via a cannula (PE 10) positioned in the ventral tail artery. An infusion line was introduced into a lateral tail vein via a 23 g needle. Electrocardiogram (e.c.g.) electrodes were placed in the dorsal skin and colon temperature was continuously monitored. Rats were placed in restraining cages, and 1 h later phenylephrine was infused at progressively increasing rates (3–50 μg kg^{-1} min^{-1}. i.v.) to elevate MAP by 40–60 mm Hg in 5–7 min (Figure 1a). The chart speed was increased each time the MAP stabilized during its ascent, and heart period (HP) was derived by averaging across 10 e.c.g. beats (Figure 1b).

The HP-MAP regression line was then constructed (Figure 1c). 'Stray points' occasionally observed at a high MAP were rejected if they fell outside the 95% tolerance limits (Figure 1c; Snedecor & Cochran, 1967). A 'guard point' (not used in the calculation) was allowed between the accepted points and the stray points, to reduce judgement error (Figure 1c).

The mean slope obtained from 66 rats was 1.19 ± 0.06 ms/mm Hg (mean ± s.e. mean) with a mean correlation coefficient of 0.91 ± 0.01. Phenylephrine had no direct effect on HP since pretreatment with atropine (1 mg/kg) + atenolol (10 mg/kg) prevented any chronotropic changes.

This method should prove useful for studying the central pharmacology of the reflex in this species. Using this method we have shown that the reflex is predominantly (but not entirely) vagal under these conditions (Redfern, Little & Stoner, 1980).

W.S.R. is an M.R.C. scholar.
We thank ICI for generous gifts of atenolol.

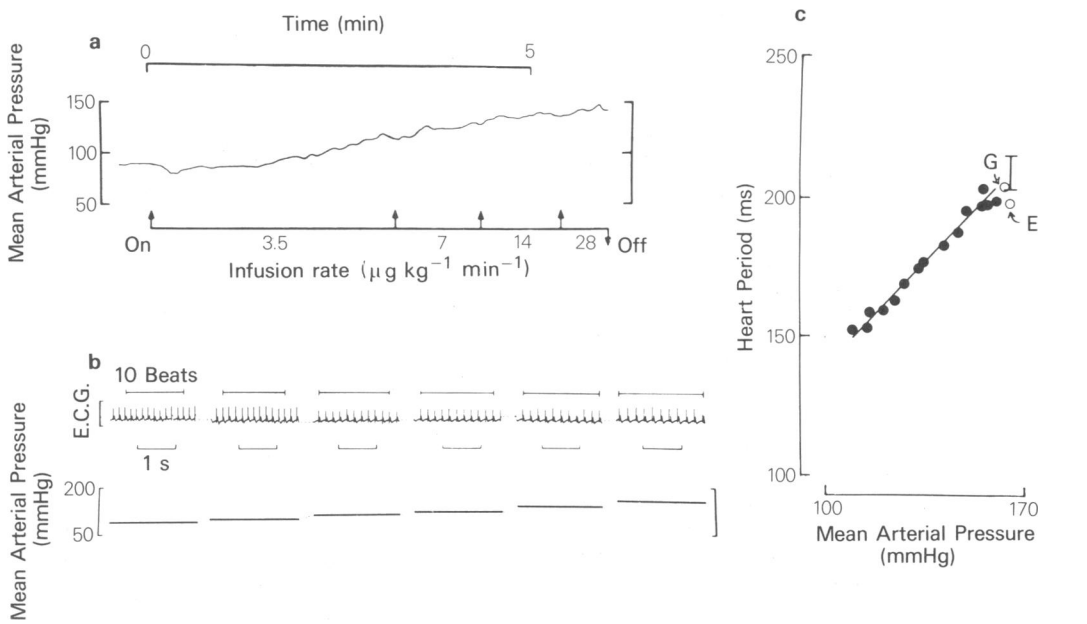

Figure 1 A. Slow speed chart record showing changes in MAP during infusion of phenylephrine. B. High speed chart recordings at 6 MAP plateaus. HP-MAP data points are obtained from at least 8 such plateaus. C. HP-MAP plot. The line of best fit was calculated by the least squares method of linear regression. The vertical bar represents the 95% tolerance limits, calculated from the residual variance about the line, with n-2 degrees of freedom. G = guard point; E = excluded point.

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An improved isolated-tissue bath suitable for automated control

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A new design of glass-constructed isolated-tissue bath is presented. The design overcomes many limitations of previous types of bath and permits easy, remote, automated control of washing cycles. Salient design points are:

1. A non-return valve which eliminates diffusion of sample drugs into the fresh bathing fluid.
2. Management of foam build-up without loss of bath fluid.
3. A vertically-folded radiator which permits viewing of tissues within the bath.
4. A removable base to the bath incorporating sintered disc and fine-control stopcock for control of gas flow.

A sketch of the aparatus is shown (Figure 1). After passage through a timer-controlled solenoid valve physiological bathing fluid enters the apparatus at inlet, C. The fluid is warmed by passing through the vertically-folded radiator, J, which is immersed in thermostatically controlled water, circulating in the outer jacket, H. The physiological fluid enters the 5 ± 0.25 ml bath past the stainless steel non-return valve, D, which closes under gravity when washing ceases.

The bath washes by overflow through five 2 mm holes, F, into the trough, E, which is drained by continuous suction from outlet I. The trough prevents vacuum variations from affecting the bath fluid level. Above holes, F, which determine the bath capacity, an outward flange, G, is provided which allows foam to collect and disperse without fluid loss.

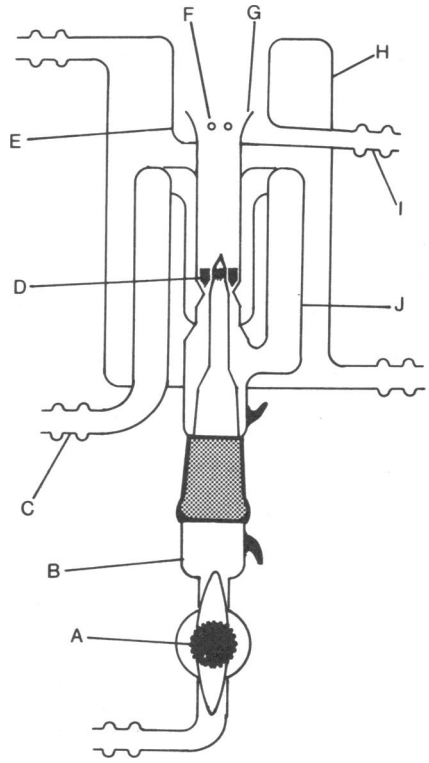

Figure 1 Design of isolated-tissue bath. See text for details.

The plug, B, incorporating a tissue attachment loop, sintered disc and gas control stopcock, A, is removable to permit easy cleaning of the apparatus.

Several baths of this design may be used simultaneously, controlled by a single timer and solenoid valve.

We thank Mr D.J. Brown and Mr D.J. Marner of our Instrument Services Unit for designing the automatic controller and stainless steel valve.

The innervated *in situ* Langendorff perfused guinea-pig heart

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The isolated Langendorff perfused guinea-pig heart suffers the disadvantages of rapid deterioration of its force of ventricular contraction and also that the sympathetic and parasympathetic components of its innervation cannot be easily removed together with the heart during isolation procedures.

The preparation to be demonstrated allows the heart and its innervation to remain *in situ* and the perfusion equipment is arranged so that it can be sited close to the animal.

The saline perfusion system consists of a glass warming coil with a jacketed outer container circulated with warm water at 40–53°C. Kreb's solution aerated with 95% O_2: 5% CO_2 is delivered to the warming coil by means of a Watson Marlow MHRE/22 peristaltic constant volume pump at a rate of 3 ml/min and the solution emerges from the coil into the glass cannula at a temperature of 37°C. Drugs are administered to the heart by means of a side arm attached to the perfusion cannula.

Male or female guinea-pigs (400–500 g) are stunned by a blow and bled from the jugular veins and common carotid arteries in the neck region. The animal is then laid on its back in a shallow dish and the thoracic cage rapidly dissected away to expose the lungs and heart. The ascending aorta is identified and a cotton ligature placed underneath by means of angled forceps and the glass tip of the perfusion cannula placed retrogradely into the cut aorta and tied. The left post-ganglionic sympathetic nerve supply is identified at the point of its emergence from the stellate ganglion and a ligature passed around it and also the left pre-ganglionic vagus nerve is identified alongside the common carotid artery.

For purposes of collection of heart perfusate the pulmonary artery is cannulated with a polythene cannula and the remaining branches of the pulmonary vessels and the vena cava are ligated.

Force of contraction of the ventricles is measured either by a tension transducer connected by a thread to the left ventricle via a pulley or by means of a small air filled balloon inserted into the cut right ventricle connected to a pressure transducer.

The preparation has been used to study the effects of sympathetic nerve stimulation on the noradrenaline output.

Rat heart ECG recording *in vitro* under normothermic and hypothermic, computer controlled conditions

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The responses of the human perfused umbilical artery preparation to histamine

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Hymenolepis microstoma in a screening test for anthelminthics

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The use of (−)N-n-propylnorapomorphine in the measurement of locomotor activity using an automatic quantitative method

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