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EXPERIMENT FIVE

Perfused Rat Hindquarters

The isolated hindquarters preparation allows examination of drug effects on resistance vessels. Responses to a range of vasoactive drugs can be clearly seen.

Physiological saline is pumped at a constant (but pulsatile) flow rate into the abdominal aorta to keep the rat hindquarters perfused. Changes in vascular resistance are recorded as changes in perfusion pressure.

Dissection

Open the abdomen of a freshly killed rat. Locate the distal colon, and tie two ligatures around it, about 1 cm apart. Cut the colon between the ligatures, and remove all the proximal bowel from the peritoneal cavity. Locate the abdominal aorta, anterior to its bifurcation, and cannulate it. With strong secateurs, cut through the rat's body.

Place the hindquarters on a metal grille over a collection dish. Gently flush physiological saline through the preparation, from a 20 mL syringe attached to the cannula, until the effluent is almost clear. Then connect the cannula to the flow-line from the peristaltic pump and heat exchanger. The pressure transducer is connected to a T-piece in the flow-line.

Experiment

Apparatus

This experiment requires a PowerLab and Bridge Amp, but uses a pressure transducer (such as the MLT 1050 or MLT0380) rather than a force transducer. Also needed are tubing and stopcocks for connecting and flushing the pressure transducer; a peristaltic pump with appropriate pump tubing, capable of supplying 3 mL/minute; and an organ bath with a heat-exchanger, to warm the saline after it has passed through the pump. (The bath itself is not used.)

Chart Settings

This is a single-channel arrangement. In Chart, the range should be chosen to suit the maximum pressure to be exerted on the transducer. The final scale after units conversion and so on should be 0 to 75 mmHg. The

sampling rate should be at least 4/s. The view compression should be 5:1 or 10:1. A low-pass filter of 10 Hz should be chosen in the Bridge Amplifier dialog box. Data Pad miniwindows can be set up to show the comment number and text, and the mean of the selection or value at the active point (using the Mean function).

Protocol

Injections of standardised volume, typically 0.1 mL, are made into rubber tubing just proximal to the cannula. Suggested concentrations for the stock solutions of vasoactive drugs are:

noradrenaline	10 µg/mL
adrenaline	10 µg/mL
isoprenaline	100 µg/mL
serotonin	20 µg/mL
dopamine	100 µg/mL
tyramine	100 µg/mL
vasopressin	0.5 U/mL (supplied in these units; 1 U ≈ 3µg)

Responses to selected agonists can be examined after changing the perfusion solution to one containing the beta-blocker propranolol (1 mg/L) or the alpha-blocker phentolamine (1 mg/L), or the catecholamine-uptake blocker desmethylimipramine (0.1 µg/L).

Physiological saline exerts no colloid osmotic pressure. In consequence, over the course of a few hours the hindquarters become visibly swollen, and there is usually a rise in basal vascular resistance.

Figure 5–1 shows the response of the isolated perfused hindquarters preparation to injections of noradrenaline and vasopressin. The regular pressure waves are due to cyclic flow from the peristaltic pump. The Data Pad Mean function has been used to quantify the pressure over several cycles, as shown in the miniwindow.

Figure 5–1

The response of isolated perfused hindquarters preparation to injection of noradrenaline (1 µg; left) and vasopressin (0.1 U; right).

