Pituitary dopamine in rats after decreased water intake

HEATHER BANNS, D. BLATCHFORD, URMA GODDEN, MARGARETHE HOLZBAUER, S.P. MANN & D.F. SHARMAN

A.R.C. Institute of Animal Physiology, Babraham, Cambridge CB2 4AT

The neuro-intermediate lobe of the pituitary gland of vertebrates is innervated by dopaminergic neurons. In the rat these neurons originate in the rostral part of the arcuate nucleus (Björklund, Moore, Nobin & Stenevi, 1973). A turnover of hypophyseal dopamine was suggested by results of experiments in which dopamine synthesis was inhibited with α-methyl-p-tyrosine (Godden, Holzbauer & Sharman, 1977). Investigations are now in progress to test a possible involvement of pituitary dopamine in the release of hormones from the gland.

In this demonstration results will be shown of experiments on rats in which a large and prolonged release of antidiuretic hormone (ADH) and oxytocin (OXY) was provoked by keeping them on a dry diet (Oxoid B41) for 72 h, followed by 1–5 days during which a 2.5% solution of sodium chloride was offered. This treatment resulted in a severe loss of Gomori positive substance from the neural lobe and the hypothalamus. A hypertrophy of the adrenal glands indicated a simultaneous increase in ACTH secretion. The dopamine content of the neurointermediate lobes of the pituitary glands from rats on a reduced water intake was approximately doubled when compared with litter-mate controls. There was also a significant rise in the catecholamine concentrations of the 'basal hypothalamus' (a structure of about 25 mm^3 including the median eminence and the arcuate, the supraoptic, and the paraventricular nuclei). There was no rise in the dopamine concentrations in the corpora striata. The reduction in water intake caused hyperaemia in the neural lobe, but not in the intermediate or the anterior lobe of the pituitary gland. Fluorescence microscopy showed a striking increase in the catecholamine-containing neurons surrounding the distended blood vessels and of non-vasomotor fibres in the posterior lobe when compared with controls.

The question whether the observed increase in pituitary dopamine concentrations is the result of the increased ADH and OXY secretion or whether dopamine plays an instrumental role in the release of ADH and OXY cannot yet be answered. It is, however, unlikely that it is the unspecific effect of an altered NaCl-water balance or of stress on dopamine-containing neurons, because the striatal dopamine content remained unchanged. In *in vitro* experiments, Bridges, Hillhouse & Jones (1976) observed a two-fold rise in the release of ADH and OXY from the superfused neurohypophysis of the rat when dopamine in a concentration of 6.5×10^{-6} M was added to the superfusion medium.

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A denervated dog's kidney preparation for studying renin release into blood and lymph

C.S. WILCOX

The Medical Unit, St. Mary's Hospital Medical School, London, W2 1NY

A method has been described previously for measuring the flow rates and composition of renal blood and lymph (Wilcox, 1976, 1977). It has been evaluated for the study of renin release.

Greyhounds are anaesthetized with pentobarbitone sodium. The experimental kidney is acutely transposed across the animal's own carotid artery and jugular vein. Lymphatic vessels are opened, lymph collected and its flow rate (LFR) measured directly. Renal venous blood is sampled through a cannula in the renal vein. A needle in the anastomosed carotid-renal artery transmits 0.154 M saline at 0.1 ml min^{-1} kg^{-1}.

Renin activity is expressed as p mole of angiotensin I generated from 1 ml of plasma or lymph during a 1 h incubation in the presence of added dog's renin substrate. Angiotensin I is measured by radioimmunoassay.

Mean values (±s.d.) from 22 experiments were as follows: renal plasma flow (RPF) was 5.57 ± 2.00 ml min kg^{-1}: LFR 4.14 ± 1.40 µl min kg^{-1}: renin activity of renal venous plasma (9.6 ± 7.2) was not different ($P>0.1$) from that of systemic arterial plasma (9.3 ± 6.4), but renin activity of lymph was invariably greater, averaging 17.7 ± 10.8. Measurements of these values were repeated after 1 h and none had changed significantly ($P>0.1$).

In 13 experiments, infusion of $MgCl_2$ solutions into the blood supplying the experimental kidney to raise its plasma magnesium concentration by 0.1 to 1.5 m mole l^{-1} increased both RPF and LFR. The renin activity of renal venous plasma rose to 12.8 ± 10.0 which was now significantly ($P<0.001$) greater than that of arterial plasma. The renin activity of renal lymph also increased significantly to 28.6 ± 9.7.

These results illustrate that with this preparation all parameters used to calculate renin release remain stable over a 1 h period. In the basal state there is nett release of renin into lymph but not into blood. It is possible to augment substantially renin release into lymph and into blood.

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Modification of transport processes across rat intestine

J. HARDCASTLE*, P.T. HARDCASTLE* & P.A. SANDFORD** (introduced by F. HOBBIGER)

*Department of Physiology, The University of Sheffield** and Department of Physiology, The Middlesex Hospital Medical School, London***

Several techniques have been developed whereby the electrical properties of the colon and the small intestine can be continually monitored. These have allowed the measurement of the effects of naturally occurring substances on electrolyte transport processes and those mechanisms dependent on the movement of ions, e.g. hexose. Changes in both absorptive and secretory events may be detected *in vivo* in response to substances introduced either into the blood stream or administered intraluminally. *In vitro* methods are used to eliminate the possibilities that the effects of administered substances are dependent on events occurring at sites away from the intestine and that the responses observed are brought about by vascular or motility changes.

An advantage of these methods is that the absorption or secretion of very small amounts of electrolytes and non-electrolytes can readily be detected, amounts which by chemical or radiochemical means are very difficult to measure, e.g. perfusion of rat ileum with 10 mM glucose at 3 ml/min for 1 min produces a marked change in potential difference (p.d.) (4.5 ± 0.4 mV–9 expts.) accompanied by a small hexose absorption (1.5 ± 0.2 µmoles, 9 expts.).

Data obtained from single experiments, following the administration of naturally occurring substances, can be employed to construct dose-response curves. Furthermore, these techniques are valuable in that they provide a tool with which the mode of action of blocking agents can be studied. Thus acetylcholine has been found to increase the p.d. across the rat proximal colon, both *in vivo* and *in vitro*, a sigmoid relationship being recorded between the change in p.d. and the log acetylcholine dose. The inability of hexamethonium and pentolinium to alter the response of acetylcholine suggested a direct effect of the transmitter substance on the colon, and the inhibitory effects of atropine suggested the acetylcholine effect is mediated by a muscarinic type of receptor. Subsequent measurements of unidirectional ion fluxes established that acetylcholine virtually abolished net Na^+ movement and induced net Cl^1 secretion. These alterations in ion movement could account for the observed electrical changes.

In a number of diarrhoeal states biologically active substances are thought to be released in excessive amounts and act by modifying absorption and/or secretory processes. For example, it has been suggested that in the carcinoid syndrome both bradykinin and 5-hydroxytryptamine may be involved in producing a net secretory state. Certainly both these substances and prostaglandins E_1 and E_2 alter the electrical properties of the intestine. The techniques demonstrated will be useful in determining the mode of action of these and other substances and how they can be inhibited.