Direct Stimulation of Adrenocortical Secretion by Synthetic Vasopressin in Dogs.* (24683)

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Administration of vasopressin to non-hypophysectomized animals and man results in an increase in output of adrenocortical steroids (1,2,3,4). This effect has been attributed to stimulation of the adenohypophysis by the vasopressin which in turn results in release of ACTH or some substance with ACTH-like activity(1-6). However, Guillemin and Hearn(7) as well as Saffran and co-workers using tissue culture technics(8) were unable to confirm any adenohypophyseal stimulation by vasopressin. More recently, Royce and Sayers(9) have noted increased adrenocortical steroid secretion in hypophysectomized as well as decapitated rats receiving vasopressin and have concluded that vasopressin must release ACTH from peripheral binding sites. Because of these somewhat conflicting reports it occurred to us that vasopressin might directly stimulate the adrenal cortex and that the adrenal perfusion technic, as developed in this laboratory(10), would be an ideal method for studying this possibility.

Methods. Normal adult mongrel dogs, weighing between 15-20 kg, were anaesthetized by intravenous administration of 0.46-0.62 mg/kg body weight of sodium pentobarbital. The hypophysis was removed by transbuccal approach(11). Adrenals were then prepared for perfusion by the technic of Hilton *et al.*(10). Donor hypophysectomized dogs were bled from femoral artery the morning of experiment. This arterial blood was perfused by mechanical pump into the adrenal preparation of recipient hypophysectomized animal. Synthetic vasopressin‡ was added to arterial circuit leading to adrenal glands at rate of 1 ml/minute for 7-10 minutes. Concentrations used varied from 0.01-0.4 pressor units/ml (0.03-1.0 μg). Blood flow rate through glands was kept constant at 10 cc/minute throughout each experiment. At end of each experiment an injection of ACTH at 1 ml/minute for 7-10 minutes and at concentration of 0.5-2.5 units/ml was given to verify adrenal viability. Adrenal venous blood was collected in graduated cylinders, rate of flow was measured, and concentration of hydrocortisone in plasma was determined by the method of Peterson *et al.*(12).

Results. Ten experiments were performed at different dosage levels of both arginine and lysine vasopressin. Control secretion rates of hydrocortisone were uniformly low, averaging less than 0.5 μg/minute, in all 10 experiments. In the lysine group (4 experiments) hydrocortisone§ secretion was increased, as much as 6-fold, over control resting rates. Following ACTH administration at end of each experiment there was further increment in hydrocortisone secretion. This same pattern of response was noted in 5 experiments in which arginine vasopressin was perfused.

An attempt was made to demonstrate a dose-response curve utilizing 0.01 unit/minute, 0.1 unit/minute and 0.4 unit/minute of arginine vasopressin (Fig. 1). A marked increase in hydrocortisone secretion was observed following initial small dose and a significant further increase was noted upon administration of 0.4 units/minute.

High doses of vasopressin appeared to result in more prolonged as well as greater hydrocortisone secretion. This can also be seen in Fig. 1, where hydrocortisone secretion was still elevated 20 minutes following end of perfusion of 0.4 unit/minute of arginine vasopressin, as contrasted with the more rapid fall off in secretion rate following 0.1 unit/minute.

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§ Spectrophotometric analysis revealed maximum absorption at wave length of 410 mμ.

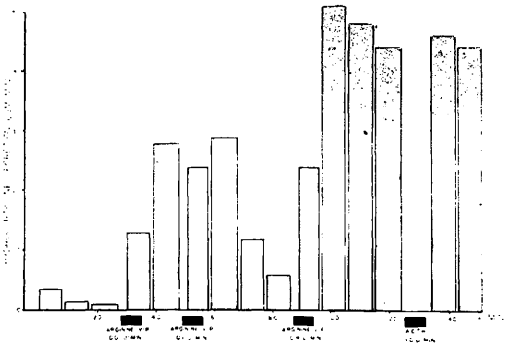

FIG. 1. Effect of graded doses of arginine vasopressin on hydrocortisone secretion from perfused adrenal glands of the dog. All substances were administered over 7 min. time interval.

The advantage of direct perfusion technic in these experiments is elimination of interference from other endocrine glands or organs. These factors may in part explain the lack of effect of vasopressin on adrenocortical secretion, noted by some investigators(13). Whole body dilution of injected vasopressin, inactivation of material by liver or at other sites prior to reaching the adrenal glands, possible species differences etc., must all be taken into account in interpretation of previously reported negative results.

The data indicate that synthetic vasopressin has a direct stimulatory effect on the adrenal glands, enhancing secretion of hydrocortisone, and appears to be in some instances as potent a stimulator as adrenocorticotrophic hormone itself.

It is evident from our studies that data which have accumulated on vasopressin stimulation of ACTH or an ACTH-like substance is not, in fact, due to such stimulation but is a direct effect of vasopressin on the adrenal cortex itself. ACTH effect in our preparation is excluded by: (1) use of synthetic vasopressin (2) use of hypophysectomized animals (3) low resting secretion rates from adrenal gland prior to vasopressin administration (4) direct perfusion of adrenal glands *via* their arterial circuit.

This surprisingly potent adrenocortical stimulating effect of vasopressin implies that it may be an important factor in reaction to stress. Our preliminary studies indicate an additive effect when vasopressin is given to ACTH-stimulated adrenal gland preparation. This raises the possibility that vasopressin stimulates adrenal steroid secretion by a mechanism which is distinct from that of ACTH.

Summary. By means of direct arterial perfusion of the adrenal glands in the dog it has been shown that synthetic vasopressin stimulates secretion of hydrocortisone. This effect is not mediated *via* the adenohypophysis or any other organ, but is rather the result of direct stimulation of the adrenal cortex by vasopressin itself.

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