PRESSINOIC ACID: A PEPTIDE WITH POTENT CORTICOTROPHIN-RELEASING ACTIVITY

by

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SUMMARY. Pressinoic acid, a synthetic hexapeptide that corresponds to the ring of vasopressin, exhibited corticotrophin-releasing activity *in vitro* in doses of 3 and 30 nanograms per ml. The related peptide, deaminopressinamide, released lesser amounts of corticotrophin in doses of 30 to 30,000 ng. Two other hexapeptides, pressinamide and deaminopressinoic acid, were devoid of corticotrophin-releasing activity at doses of 30 to 30,000 ng.

The existence of a hypothalamic agent for the control of the release of corticotrophin from the adenohypophysis was clearly enunciated by Harris in 1955 (1). The presence of corticotrophic-releasing activity in extracts of the hypothalamus and the neurohypophysis was demonstrated in 1955 by *in vitro* assays by Saffran, Schally and Benfey (2) and by Guillemin and Rosenberg (3). In subsequent years several attempts were made to purify the corticotrophin-releasing factor (CRF) in the extracts, but only partial purification was achieved (4-7). Recently, other hypophysiotrophic hormones of the hypothalamus have been identified, including those responsible for the control of the release from the pituitary gland of TSH (8,9), LH and FSH (10), MSH (11,12) and G.H. (13). All of these are peptides, with 3 to 10 amino acid residues and many contain the pyroglutamyl residue.

The partial structural similarity of corticotrophin and MSH, and the simultaneous presence of both hormones in the intermediate lobe of the

TABLE I. Corticotrophin-Releasing Activity of Vasopressin Ring Peptides

Peptide	Dose (ng./ml.) 3	30	300	3,000	30,000
	Activity as % of 300 ng. Lysine Vasopressin				
Deaminopressinamide		14	26	68	76
Pressinoic acid	25	170	0	0	0
	26	121	0		
		134			
Deaminopressinoic acid		0	0	0	0
Pressinamide		0	0	0	0

Corticotrophin-releasing activity of vasopressin ring compounds, expressed as percentage of corticosterone formed by the corticotrophin released from a rat anterior pituitary lobe in vitro by 300 ng. of synthetic lysine vasopressin (Sandoz). Single halved pituitary glands of female CFE rats weighing 100 to 150 g. (Carworth, Portage, Mich.) were preincubated for two hours in Krebs-Ringer-bicarbonate-glucose (2%) medium under 5% CO_2 in O_2, and then for one hour periods each with 300 ng. lysine vasopressin and the sample of vasopressin ring peptide. The incubations were separated by a one hour wash period. The media from the pituitary incubations were added to rat adrenal quarters superfused according to Saffran et al. (17) for estimation of the steroidogenic response to the corticotrophin released. The steroidogenic effect of the unstimulated release of corticotrophin by the pituitary tissue and the possible steroidogenic effect of the peptide directly on the adrenal tissue were measured together by adding the peptide to the media from pituitary tissue incubated without peptide for one hour.

In 13 assays, the standard dose of 300 ng. of vasopressin released sufficient corticotrophin to increase the formation of corticosterone to 185±10 (s.e.) % of the amount formed with the control pituitary.

Vasopressin and some preparations of CRF have yielded inverted U-shaped dose-response curves, in which high doses were without activity (18,19).

pituitary gland (14), suggested to us that analogous control mechanisms may operate for both hormones. The peptides that control the release of MSH have been identified with the tail and ring portions of oxytocin, with inhibitory (11) and stimulatory (12) activities respectively. Therefore, we tested compounds representing the ring portion of vasopressin for corticotrophin-releasing activity.

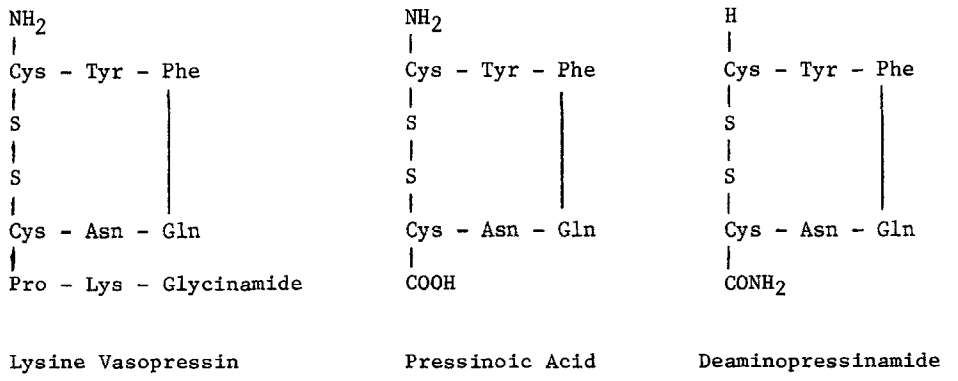

Figure 1. Structures of lysine vasopressin, pressinoic acid and deaminopressinamide.

Recently, four peptides related to the ring of vasopressin were synthesized by Ferger, Jones, Dyckes and du Vigneaud (15). These were devoid of pressor activity. We tested the peptides for corticotrophin-releasing activity by minor modification of the method of Saffran (16), using lysine vasopressin as a standard. One of the peptides, pressinoic acid (Fig. 1), was active in the range of 3 to 30 nanograms per pituitary gland (Table I). Higher doses were inactive. Deaminopressinamide was active at doses of 30 to 30,000 nanograms. Pressinoic acid released more corticotrophin at optimal doses than did deaminopressinamide. The two other ring peptides, deaminopressinoic acid and pressinamide, were inactive at the doses tested.

Pressinoic acid was devoid of corticotrophic activity in vitro in the system of Saffran, Matthews and Pearlmutter (17).

Previously, the hormones of the posterior pituitary gland, vasopressin and oxytocin, and several of their analogues were the only well characterized substances that exhibited corticotrophin-releasing activity (18). However, none of these was equal in potency to the CRF isolated from neurohyphophysial and hypothalamic tissues, which was active in nanogram amounts (4). Pressinoic acid has corticotrophin-releasing activity in this range.

The fact that pressinoic acid exhibits corticotrophin-releasing activity at a low dose level in vitro does not establish its identity as the physiological CRF. However, the activity of pressinoic acid should furnish a clue to the identity of CRF. Moreover, the absence of pressor activity in pressinoic acid, coupled with its ability to stimulate the release of ACTH from the pituitary, makes it an almost ideal candidate for use in testing pituitary ACTH reserves and the capacity of the pituitary to respond to CRF.

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