

Inhibition by Oxytocin of Vasotocin-Induced Activities in Toads. (27495)

C. BARKER JØRGENSEN AND LIS OLESEN LARSEN

(Introduced by J. W. Everett)

Laboratory of Zoophysiology A, University of Copenhagen, Denmark

The antidiuretic effect of vasopressin in the toad *Bufo bufo*(1) and of lysine oxytocin in the frog *Rana esculenta*(2) may be inhibited by oxytocin. So far, vasopressin has not been demonstrated in amphibians; and lysine oxytocin is a synthetic analogue of the chemically known neurohypophysial octapeptides and unknown from the animal kingdom. The amphibian neurohypophysis contains vasotocin and probably oxytocin(3,4,5). It was therefore of interest to see whether oxytocin may also inhibit the water balance activities of vasotocin in amphibians.

Methods. The experiments were made on males of the toad *Bufo bufo* (L.) weighing some 25-50 g. The toads were kept in tap water; cutaneous water uptake and diuresis were measured as described previously(6). Highly purified vasotocin* and synthetic oxytocin (Syntocinon Sandoz) were injected subcutaneously. The potency of the preparations was expressed in international units; the absolute amounts of the hormones have been estimated by means of the conversion factors suggested by Boissonnas *et al.*(7).

Results. It appears from Table I that injection of 0.6 or 6 m μ g of vasotocin strongly reduced urine flow; this antidiuretic effect was prevented by simultaneous injection of equal amounts of oxytocin, whereas one-tenth the amounts of oxytocin was ineffective. It is noteworthy that in larger amounts, 70 and 700 m μ g, oxytocin itself acted antidiuretically. Furthermore, 6 m μ g vasotocin slightly increased the rate of water uptake through the skin; equal amounts of oxytocin may also inhibit this effect of vasotocin. Larger amounts of oxytocin itself enhanced the cutaneous water uptake.

Discussion. Attempts have often been made to clarify neurohypophysial function in amphibians by studying the water balance and

other effects of injections of crude neurohypophysial extracts that may have contained oxytocin besides one or more other neurohypophysial octapeptide hormones. The present findings show that the results from such experiments may be misleading as regards the presence and potency of active principles in the amphibian neurohypophysis. Small amounts of extract of the neurohypophysis of the toad *Bufo bufo*, but not of the frog *Rana temporaria*, were thus found to produce anti-diuresis when injected in toads or frogs(1). It now seems that the result does not necessarily imply that the neurohypophysis of the frog contains no strongly acting antidiuretic principle; the result might be due to a lower ratio of vasotocin to oxytocin in the frog than in the toad.

For vasotocin to be of importance in the normal water balance, it would seem necessary for amphibians to be capable of selective release of this hormone from the neurohypophysis.

Summary. In the toad *Bufo bufo* (L.), the antidiuretic effect of 0.6 or 6 m μ g vasotocin was prevented by simultaneous injection of

TABLE I. Effect of Oxytocin on Vasotocin-Induced Anti-Diuresis and Cutaneous Water Uptake in *Bufo bufo*.

Doses inj./toad in m μ g:			
Vasotocin + Oxytocin		Diuresis	Water uptake
.6	+	0	33
		.07	23
		.7	103
		7	73
6	+	0	13
		.07	3
		.7	10
		7	87
0	+	7	109
		70	43
		700	1

Diuresis and water uptakes are expressed in % of values after inj. of 0.1 ml water; values are avg of 7 to 14 single determinations. The t-value of the difference between water uptakes after inj. of 6 m μ g vasotocin and of 6 m μ g vasotocin + 7 m μ g oxytocin was 2.6, and $P = 0.02$.

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equal amounts of oxytocin. The slight increase in cutaneous water uptake produced by 6 m μ g vasotocin was reduced by simultaneous injection of the same amount of oxytocin. As the amphibian neurohypophysis apparently contains both vasotocin and oxytocin, attempts to elucidate amphibian neurohypophyseal function by studying the effects of injections of non-fractionated neurohypophyseal extracts may provide misleading results as regards presence and potency of neurohypophyseal hormones.

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Effects of Hormone Administration on Collagen Biosynthesis in the Rat.* (27496)

L. A. BAVETTA, I. BEKHOR AND M. E. NIMNI

*Department of Biochemistry and Nutrition, School of Dentistry,
University of Southern California, Los Angeles*

We recently reported on the ineffectiveness of methyl testosterone as well as growth hormone in counteracting the catabolic effects of methyl prednisolone on nitrogen metabolism and body weight(1). Nevertheless, growth hormone in presence of methyl prednisolone (Medrol)[†] partially prevented the inhibitory effect of the former on collagen biosynthesis around the subcutaneously implanted polyvinyl sponges. To understand further the mechanisms involved in the interaction of different hormones on these processes, we investigated the combined effects of methyl prednisolone and stilbestrol on rate of collagen biosynthesis. Simultaneous addition of androgens (methyl testosterone) was also investigated for this purpose.

In connection with our previous findings that growth[‡] hormone partially prevented the inhibitory effect of methyl prednisolone on collagen formation we decided to further in-

crease the level of this hormone to try to enhance this protective effect. Simultaneously with the specific studies on collagen, rate of synthesis of other tissue proteins was also investigated. Weight changes and rate of uptake of radioglycine by the perineal muscles were studied. The rate of development of these muscles, particularly the levator ani, has been very frequently interpreted as a measurement of the anabolic activity throughout the organism(2,3), particularly in reference to anabolic activity of androgenic steroids.

Methods. The experimental procedure was similar to that previously described(1). Young growing male Holtzman rats of average initial weight 160 to 170 g were used. The animals were implanted subcutaneously with sterile polyvinyl sponges (Ivalon) and fed the following diets: A basal diet (control) containing 24% vitamin-free casein, 61% corn starch, 10% corn oil, 5% of Wesson salt mixture, .2% choline HCl and an adequate supply of vitamins. 6-Methyl prednisolone (20 mg/kg diet), methyl testosterone (20 mg/kg diet) and stilbestrol (3 mg/

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