Comparison of Pharmacological Effects of Lysine and Arginine Vasopressins.* (22300)

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The neurohypophyseal octapeptides, oxytocin and vasopressin, have been separated from the posterior lobes of oxen as highly purified substances by du Vigneaud and his collaborators(1,2). Related studies of hog pituitary glands led to the isolation of an oxytocin identical in all respects with the oxytocin of the ox(1). Hog vasopressin, however, differs from ox vasopressin in one respect: the arginine of ox vasopressin is replaced by lysine(3). The latter hormones are therefore conveniently designated as arginine or lysine vasopressin according to the amino acid characteristic of each. The pharmacological effects of the two vasopressins differ in one interesting respect which is described in this report.

Material and methods. Two preparations containing lysine vasopressin of different degrees of purity were used: one, a natural lysine vasopressin, purified but not regarded as the pure hormone (No. 686) and the other, an extract of acetone-desiccated hog posterior lobes (No. 700). The purified natural lysine vasopressin was prepared in the Department of Biochemistry of Cornell University Medical College; we are indebted to Prof. V. du Vigneaud for the gift of this preparation. The extract of hog posterior lobe powder was made by suspending the powder in dilute acetic acid at pH 3.0, boiling for 1 minute and filtering after cooling. The solutions of this extract and of the purified natural lysine vasopressin, also dissolved in aqueous acetic acid solution at pH 3.0, were sealed in ampoules and immersed in boiling water for 10 minutes before final storage in the refrigerator at 1.0°C. The vasopressor potency of the stored solutions in terms of U.S.P. standard were as follows: No. 686, 1.34 units per ml, and No. 700, 3.54 units per ml. The absolute potency of the samples at the time of use were 134 units per mg for No. 686, and 1.35 units extracted from 1 mg of powder for No. 700. The methods of assay have already been described(4). They employ intravenous injections in the anesthetized rat (vasopressor response) and lactating rabbit (milk-ejecting response) and in the trained unanesthetized hydrated dog (antidiuretic response). The antidiuretic response was also determined after subcutaneous injection in the hydrated unanesthetized rat by the method of Burn (5). Lastly, the avian depressor response to intravenous injection was measured in the anesthetized fowl by Coon's method(6).

Results. Because the vasopressor assay was considered the most convenient and accurate to perform in terms of expenditure of time, the potency as found by this method was chosen as the standard of comparison with the other pharmacological properties. The pattern of the pressor response to lysine vasopressin with regard to dose-response relationship and duration of action was identical with that elicited by the U.S.P. reference standard or by arginine vasopressin. The variability in potency from ampoule to ampoule is indicated by the following result: 134.3 units per mg with a standard error of ± 5.6 units for No. 686 from vasopressor assays in 12 rats with as many ampoules.

In Table I, the relative potencies of natural arginine and lysine vasopressins compared by the different tests are listed. In addition, the potencies of the extract of hog posterior pituitary powder are recorded; this extract also contained oxytocin which accounts for the high potency of this extract as tested for its effects on the blood pressure of the fowl and

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TABLE I. Relative Potency of Arginine and Lysine Vasopressins.

Preparation	Tests of potency				
	Pressor† (rat, i.v.)	Antidiuretic (Dog, i.v.)	Antidiuretic (Rat, s.c.)	Avian depressor (fowl, i.v.)	Milk-ejecting (rabbit, i.v.)
Arginine vasopressin (natural)	100	100	100	14	17
Lysine vasopressin (natural)	100	15	110	16	22
Extract of hog post. pituitary*	100	17	88	127	130

* Contained both vasopressin and oxytocin.

† In these experiments, the pressor effects of the 3 preparations in the rat were arbitrarily assigned a value of 100.

on the pressure in the lactating mammary gland of the rabbit. The avian vasodepressor and milk-ejection effects of lysine vasopressin are qualitatively indistinguishable from those of arginine vasopressin. Also the avian depressor-pressor and milk-ejection-pressor ratios show within the limits of experimental error a satisfactory agreement. In contrast to the findings in the above mentioned bioassays, the relative antidiuretic potency of lysine vasopressin after intravenous injection in the dog is much lower than that of its arginine congener and requires more detailed consideration.

Whenever highly purified lysine vasopressin is administered intravenously to the unanesthetized hydrated dog, the antidiuretic response differs qualitatively as well as quantitatively from that observed following the injection of arginine vasopressin. First, the urine flow in the first 5-minute period following the intravenous injection of lysine vasopressin is about the same as in the second 5-minute period, whereas the antidiuresis produced by the arginine compound is commonly much more marked in the second period (Fig. 1). Secondly, lysine vasopressin as assayed intravenously for its antidiuretic action in the dog, has about one-sixth of the potency of arginine vasopressin when both are compared with their vasopressor potencies in the rat (Table I).

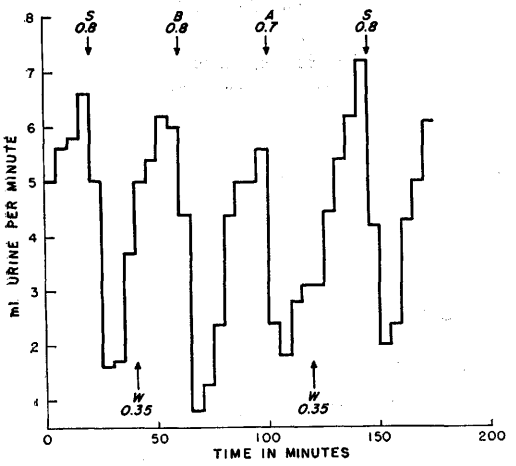

FIG. 1. Antidiuretic response in trained unanesthetized dog after intrav. injections of: S. U.S.P. Posterior Pituitary Reference Standard, 1 mU/ml. B. Extract of human infundibular stem, No. 722 (containing arginine vasopressin), 1:20. A. Purified lysine vasopressin, No. 686, 6.7 mU/ml according to vasopressor assay in the rat. W represents fractions of liters of water by stomach tube in addition to 0.4 liter at −75 and −60 min.

In an endeavor to compare the antidiuretic potencies of the vasopressins by a method employing a different route of administration, assays were performed in the rat by the method of Burn. Eleven groups of 4 animals each were employed using 2 dose levels of each preparation in at least 10 groups (40 rats). The parameter of time to 50% excretion of the water load was chosen since it appeared to be less influenced by the retention of urine in the bladder. Under these conditions, lysine vasopressin, like its arginine relative, evokes an antidiuresis which is equivalent to its pressor potency. To learn whether there is a general difference in the response of the dog and rat to lysine vasopressin, quantitative estimates of the vasopressor potency of the hormone after intravenous injection were made in the dog. The vasopressor action of lysine vasopressin was identical in the dog and rat in terms of U.S.P. standard.

Discussion. Experiments with highly purified arginine vasopressin demonstrated that the antidiuretic hormone is probably identical

with the vasopressor hormone of the neurohypophysis of oxen. In the quantitative determination of arginine vasopressin in an extract either type of assay could be employed. With the isolation of another naturally-occurring vasopressin which has a 1:6 murine vasopressor-canine antidiuretic ratio of doses, assays for antidiuretic hormone must take into account the source of the preparation, the species used for assay and the route of administration. The low potency of lysine vasopressin as an antidiuretic agent in the dog could be attributed to a difference in the response of another species. This explanation is untenable as a general statement because the vasopressor potency of lysine vasopressin, in comparison with the U.S.P. standard, is the same in the rat and dog.

Other investigations, some of which are still in progress, indicate that extracts of the neurohypophysis of man, the monkey, dog, rat, ox, sheep and camel, when assayed by intravenous methods, show a 1:1 ratio for vasopressor and antidiuretic potencies. The work described in this paper suggests that arginine vasopressin is the principal if not the only neurohypophyseal hormone responsible for these actions in the above mentioned species.

Summary. 1. The pharmacological effects of lysine and arginine vasopressins are the same with respect to their relative vasopressor, milk-ejecting and avian depressor potencies. 2. The ratio of pressor potency to antidiuretic potency is 1 for arginine vasopressin and 6 for lysine vasopressin when the antidiuretic potency is determined by intravenous injection in the hydrated, unanesthetized dog. On the other hand, this ratio of potencies is 1 for both vasopressins when the antidiuretic potency is determined by subcutaneous injection in the hydrated, unanesthetized rat. 3. Lysine vasopressin, compared with the U.S.P. standard or arginine vasopressin, is equally potent as an intravenous pressor agent in the dog and rat. Potency refers to *relative* potency; potency in units per mg is much higher for purified arginine vasopressin. 4. Lysine vasopressin is peculiar to the hog. The vasopressin of man, the macaque monkey, dog, rat, ox, sheep and camel appears, by pharmacological criteria, to be arginine vasopressin. These conclusions are tentative and must be confirmed by the isolation of the hormone from each species, other than the ox and hog, together with the demonstration of the amino acid composition of each vasopressin.

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Arterial Pressures in Tourniquet Shock. (22301)

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In this study arterial pressure was recorded during the entire period of tourniquet shock in the rat. The object was to correlate the pressure with the course and fatal outcome of the shock state.

Method. Our standard method for the production of shock in rats involves a high unilateral tourniquet applied to the left hind extremity for 5 hours. The technic was described previously(1). White male rats weighing about 250 g were used. Prior to removal of the tourniquet, the animals were anesthetized with 2% Nembutal, given intraperitoneally, and each received 0.2 cc liquid