Histamine as an Extremely Potent Releaser of Vasopressin in the Rat

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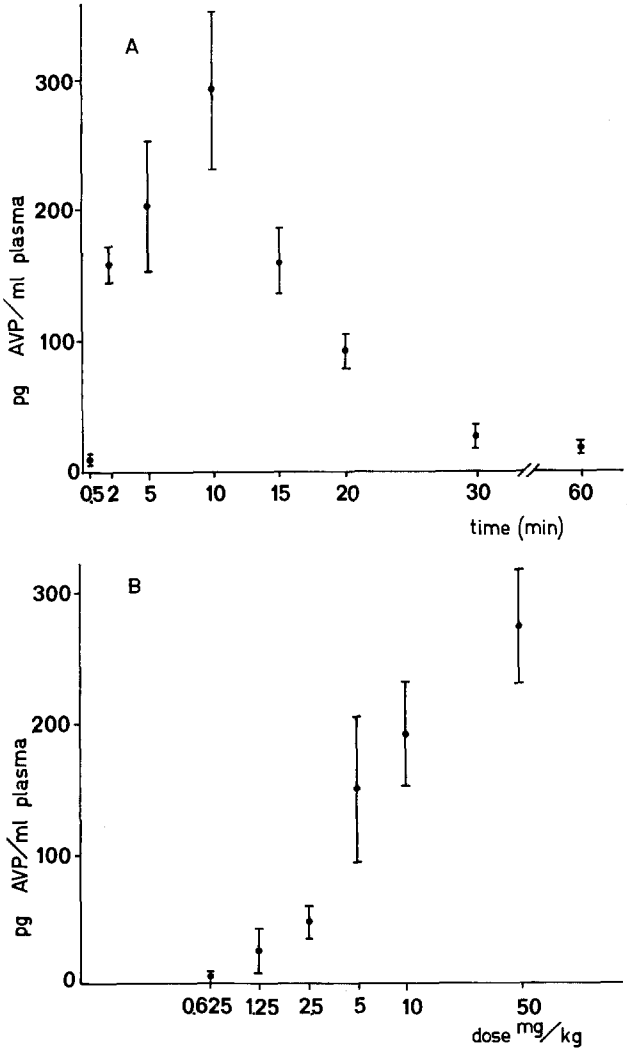
Summary. Intraperitoneal and intraventricular injection of histamine induces a very fast and high elevation of vasopressin in rat plasma as determined by radioimmunoassay. The effects are dose and time related. The intraventricular injection is more effective with regard to time and dose than the intraperitoneal injection.

Histamine may play a role as a neurotransmitter in the hypothalamus[2]. It is present in relatively high concentrations in this area of the brain of the Rhesus monkey, in particular in the supraoptic nucleus (SON). The posterior lobe of the pituitary contains considerable amounts of histamine as well[3]. In addition, intracerebroventricularly (i.c.v.) administered histamine causes antidiuresis in the cat[4] and antidromically identified supraoptic neurones can be excited by local application of histamine[5].



A) Time relationship for the rise of the vasopressin level in rat plasma after i.p. injection of histamine acid phosphate in a dose of 50 mg/kg body wt. The vertical bars show standard error of the mean ($n = 5$). B) Dose-response relationship for the rise of vasopressin levels in rat plasma after i.p. injection of histamine acid phosphate. The samples were collected by decapitation 10 min after the injection. The vertical bars show standard error of the mean ($n = 5$).

Central effects of histamine on drinking and on avoidance behaviour of rats have also been reported[6]. Several studies suggest that histamine injection is associated with the release of vasopressin[2], but direct proof, by measuring the vasopressin concentration in the plasma, has never been provided. The present studies were therefore designed to measure the arginine-8-vasopressin (AVP) levels in plasma of rats after i.p. administration of histamine in relation to time and dose. In addition, histamine was administered i.c.v. in various doses and plasma AVP levels were measured 2 min and 10 min after the application to investigate the possibility of a physiological role of histamine in the release of vasopressin from the posterior pituitary.

Male rats, weighing 150–200 g, of an inbred Wistar strain (CPB-TNO, Zeist, The Netherlands) were housed 5 to a cage on sawdust and fed ad libitum food and water. The animal house was illuminated from 05.00 h to 19.00 h. Histamine acid phosphate (B.D.H.) was injected i.p. using 0.1 ml 0.9% NaCl as vehicle.

I.c.v. injection of histamine was performed in a volume of 1 µl via an indwelling polyethylene cannula into the lateral ventricle. The operation was performed under light ether anaesthesia and the animals were allowed to recover from the operation for 4 days. At the end of each experiment, the position of the tip of the cannula was localized by macroscopical inspection of the formalin fixated brains.

Blood samples were obtained between 09.00–11.00 h by decapitation at 30 sec, 2 min, 5 min, 10 min, 15 min, 20 min, 30 min and 1 h after the i.p. injection. Blood sampling following i.c.v. injection occurred at 2 min or at 10 min after administration. The blood was collected in cooled heparinized polypropylene tubes.

Plasma vasopressin levels were measured by the use of a radioimmunoassay for AVP[7,8]. The hormone was extracted from the plasma with activated (3 times overnight at 700 °C) Vycor glass powder (Corning Glass Works, Multiform Prod., New York, U.S.A.). Plasma of Brattleboro rats, homozygous for diabetes insipidus

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which lack endogenous vasopressin[9], served as a control for aspecific effects. Cross reactivity of the radioimmunoassay with oxytocin was $< 0.1\%$. The assay reliably detects 0.5 pg AVP/ml. Mean recovery of standard AVP from plasma was 69.4 $\pm$ 6.5% ($n = 167$). The data are not corrected for recovery. Statistical analysis of the data was performed by using Student's t-test.

The basal level of vasopressin in plasma of Wistar rats as measured by radioimmunoassay was 1.2 $\pm$ 0.3 pg/ml ($n = 9$). No vasopressin was detectable in plasma of Brattleboro rats, homozygous for diabetes insipidus.

Figure A shows the rise in AVP concentration in rat plasma after i.p. injection of 50 mg histamine per kg body wt. A level of 10.3 $\pm$ 3.7 pg/ml was reached within 30 sec, which increased to 291.3 $\pm$ 61.1 pg/ml after 10 min. 1 h after administration of histamine, the concentration of AVP was still augmented (17.3 $\pm$ 4.5 pg/ml).

Figure B shows the plasma AVP concentration in samples collected 10 min after the i.p. injection of graded doses of histamine. The lowest dose used, 0.625 mg/kg body wt., caused a small but significant rise ($p < 0.05$) in plasma AVP concentration to 3.6 $\pm$ 1.2 pg/ml, while the highest dose, 50 mg/kg body wt., elicited a rise which was of the same magnitude as found in the first experiment (Figure A): 272.0 $\pm$ 44.0 versus 291.3 $\pm$ 61.1 pg/ml.

When histamine was administered i.c.v. in a dose of 0.6 mg/kg body wt., a rise to 34.9 $\pm$ 7.0 pg/ml ($n = 4$) after 2 min was measured. 8 min later this value had fallen to 2.7 $\pm$ 0.2 pg/ml ($n = 5$). A level of 66.5 $\pm$ 19.2 pg/ml was found 2 min after i.c.v. administration of 3 mg/kg body wt.

The results clearly indicate that histamine is a potent releaser of vasopressin in rats as determined by radioimmunoassay. A direct relationship between the plasma vasopressin concentrations and the dose of i.p. injected histamine was found. Results from experiments in progress in our institutes indicate that neither ether stress, nor emotional stress, nor dehydration are able to induce such an increase of vasopressin secretion into the peripheral blood[8]. Moreover, the vasopressin levels in the plasma appeared to rise very quickly after injection of histamine. I.c.v. administration of histamine was even more effective in releasing vasopressin into the peripheral circulation, with regard to time and dose, than i.p. injection of the drug. These results therefore support the view that histamine may have a central effect on the release of vasopressin. Whether histamine acts directly on the vasopressin secreting neurones, or via adrenergic fibres involved in the release of this peptide[10], remains to be elucidated.

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Successive Clutches Induced by Surgical Excision of Post-Ovulatory Oocytes in the Lizard *Cnemidophorus uniparens*

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Summary. 19 animals had eggs excised from the oviducts soon after ovulation. Number of clutches was nearly tripled in excised animals as compared to controls. An influence of eggs in the oviduct on number of clutches is suggested and may indicate a neuronal link between oviduct and hypothalamus.

Although a wealth of data exists in reptiles regarding annual reproductive cycles[3–6], reproductive strategies[7], and reproductive physiology[5, 6, 8–14], little is known regarding the endocrine control of ovulation[9, 10, 15, 16]. Further, there are few if any studies dealing with the endocrine or neuronal effects of the presence or absence of eggs in the oviduct on the time interval between clutches. This report deals with the possible influence of oviducal eggs on the time interval between clutches in *Cnemidophorus uniparens*.

Four groups of animals were used in this analysis. 21 were collected during 6–7 July 1967, 8 during 21–23 July 1968, 5 during 21–22 June 1972 and 7 during 6–10 June 1973. The 1st, 2nd and 4th groups were collected in Socorro County, and the 3rd in Sierra County, New Mexico (approximately 25 miles apart).

They were maintained at a preferred temperature of 33–36 °C during the light and at 20 °C during the dark period. All animals were exposed to a 10L:14D period. Animals were housed in groups of 2 to 3 in 15 gallon terraria (60 cm long) containing a 2 cm sand substrate. Illumination and heat were provided by 125 GE infra-red reflector lamps.

A total of 19 females of the first 2 groups had their eggs excised from the oviducts as part of a separate study[17] immediately after the time of ovulation. The egg excision process was repeated for any animal that underwent a successive clutch. All others ($n = 22$) were not laparotomized and were used as untreated controls. Students' t-test was used for statistical comparison.

Individuals of all groups ovulated during a 5 month interval between December and April, corresponding to the breeding season described for this species in the laboratory[17]. The mean number of clutches per individual was significantly higher ($p < 0.005$) in the experimental groups (3.00 $\pm$ 0.20) than in the control groups (1.32 $\pm$ 0.10). Similarly, the percent of individuals having more than 2 clutches was significantly higher ($p < 0.005$) in the experimental than the control group (68.0 and 0.00% respectively). As many as 4 to 5 clutches were layed during one reproductive season in animals that survived the laparotomies. 6 animals ovulated twice, 8 ovulated 3 times, 4 ovulated 4 times and 1 animal ovulated 5 times. Of the controls 15 developed 1 clutch and 7 developed 2.

These results demonstrate for the first time a possible neuronal influence of the presence of eggs in the oviduct on number of clutches in a reptile and may indicate a neuronal connection between oviducts and hypothalamus. A similar neuronal link between oviducts and hypothalamus has been suggested for the hen[18]. It was found that the presence of an irritant (thread loops) in the magnum