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J. BUGAJSKI, A. GĄDEK-MICHALSKA, A. OŁOWSKA, J. BORYCZ, R. GŁÓD

CENTRAL HISTAMINERGIC MECHANISMS MEDIATE THE VASOPRESSIN-INDUCED PITUITARY ADRENOCORTICAL STIMULATION

Department of Physiology, Institute of Pharmacology, Polish Academy of Sciences, Cracow, Poland

Involvement of histamine receptors and hypothalamic and hippocampal histamine in stimulation of the hypothalamic-pituitary-adrenal (HPA) axis by vasopressin (AVP) was investigated in conscious rats. The HPA activity was assessed by measuring serum corticosterone levels. One hour after administration AVP, (5 μg/kg) given ip significantly raised the serum corticosterone and hippocampal histamine levels, while the hypothalamic histamine content was not affected. Pretreatment with the inhibitor of the brain histamine synthesis α-fluoromethylhistidine (α-FMH) (50 mg/kg ip) considerably reduced both the AVP-elicited serum corticosterone response and the hypothalamic and hippocampal histamine levels. The histamine H_1- and H_2-receptor-antagonists mepyramine (0.01 mg/kg) and ranitidine (0.1 mg/kg), given ip 15 min prior to AVP, significantly impaired the AVP-induced rise in the serum corticosterone level and totally abolished the AVP-elicited increase in the histamine content in the hippocampus; moreover mepyramine significantly lowered this content in hypothalamus. Pretreatment with the histamine H_3-receptor antagonist thioperamide (5 mg/kg ip) also significantly decreased the AVP-elicited corticosterone response, but did not alter the histamine content in either brain structure examined. These results indicate that central histamine H_1-, H_2- and H_3-receptors significantly mediate the stimulatory action of AVP on the pituitary-adrenocortical axis. Hippocampal histamine may be involved in mediation of the AVP-induced effect *via* H_1- and H_2-receptors. The inhibitory effect of thioperamide seems to be located directly at non H_3-intracellular sites of the pituitary-adrenocortical axis.

Key word: *Vasopressin, pituitary-adrenocortical axis, corticosterone, histamine receptor antagonists, α-fluoromethylhistidine.*

INTRODUCTION

Apart from its well-known pressor and antidiuretic effects (1), vasopressin, a peptide hormone sythesized in the hypothalamic supraoptic and paraventricular nuclei (PVN), significantly contributes to activation of the

pituitary-adrenal system, particularly under stress circumstances (2—4). Although vasopressin was the first identified secretagogue of adrenocorticotrophin (ACTH), CRH is now generally regarded as the primary and most potent activator of the pituitary-adrenocortical axis (5). The co-existance of AVP and CRH in around a half of the CRH axons in the external zone of the median eminence in normal rats has been established (6, 7). Vasopressin and CRH are secreted into the hypophysial portal blood from axon terminals projecting to the external zone of the median eminence. On reaching the anterior pituitary corticotrophs, both these peptides act synergistically to stimulate the release of ACTH, which in turn, stimulates the synthesis and release of glucocorticoids from the adrenal cortex. We have recently shown that after systemic administration, AVP is, on a molar basis, almost as potent as CRH in stimulating of the pituitary-adrenocortical response (8).

The release of both CRH and AVP from the hypothalamic neurons, and of ACTH from the anterior pituitary corticotrophs is coregulated by neurotransmitters at the hypothalamic and/or pituitary level (9—11).

Besides the hypothalamus, also the hippocampus is known to be involved in regulatory mechanisms of the HPA axis (12—15) and evidence has been accumulated in support of the role of histamine as a neurotransmitter in this structure (16, 17). All regions of the hippocampus receive a histaminergic input, and histamine has a facilitatory influence on the hippocampal excitatory system.

Reports on the role of histamine in AVP release are contradictory. When given systemically to conscious rats or icv to conscious goats, histamine increased the plasma concentration of AVP (18, 19). Administered to the PVN/anterior hypothalamus region, histamine stimulated the AVP secretion *via* a local release of noradrenaline and α_1-adrenoreceptors activation (20).

However, possible involvement of histamine and its receptors in the AVP-induced ACTH and corticosterone secretion is not known.

The aim of the present study was to determine whether histamine H_1-, H_2- and H_3-receptors and hypothalamic and hippocampal histamine are involved in the vasopressin-induced stimulation of ACTH and corticosterone secretion in conscious rats.

MATERIALS AND METHODS

Adult male Wistar rats weighing 190—220 g which had free access to food and water, were used in all experiments. The animals were kept in groups of 7 per cage under standard laboratory conditions on a diurnal light cycle at least one week prior to the experiment. The rats were arbitrarily assigned to one of the experimental groups. The indicated doses of drugs were injected intraperitoneally in a volume of 1 ml/kg. Control rats received 0.2 ml of saline. Histamine receptor

antagonists were administered 15 min, and α-FMH 2 h before vasopressin. One hour after the last injection, the rats were decapitated, their trunk blood was collected, and hypothalami and hippocampi were isolated. Control rats were decapitated concurrently with the experimental group. After centrifugation, serum aliquots were frozen until the assay. The serum corticosterone concentration was determined fluorometrically and expressed as μg per 100 ml. One analysis was performed in each rat's serum, but 6 animals were used for each data point. All experiments were performed between 9 and 12 a.m., and all decapitations took place between 11 and 12 a.m. to avoid corticosterone level fluctuations due to the diurnal rhythm.

For histamine determinations, the rats were decapitated at the required time, their brains were quickly removed, placed on glass kept on ice, and washed with ice-cold saline. The hypothalami and hippocampi were isolated and stored at −80°C until the assay. A 10 or 20% (w/v) tissue homogenate was prepared in 0.4 M perchloric acid, the homogenate was then centrifuged and the supernatant was adjusted to pH 5—6 with 0.2 M KOH. A 0.5 ml aliquot was passed through a Cellex P column (5 × 30 mm) and washed sequentially with 0.5 ml of 0.03 and 0.1 M sodium phosphate buffer (pH 6.2). Histamine was eluted with 1.5 ml of 0.07 M hydrochloric acid and, after condensation with o-phtaldialdehyde, it was estimated fluorometrically at 360/450 mm.

The drugs used were: arginine vasopressin, rantidine hydrochloride (Sigma), mepyramine maleate (May and Baker), thioperamide maleate (Tocris Cookson), and α-FMH (a gift from Dr. Kollonitsch, Merck Sharp and Dohme).

The results were calculated as a group mean ± SEM. Statistical evaluation was performed by an analysis of variance, followed by individual comparisons by Duncan's test.

RESULTS

Vasopressin-induced changes in the serum corticosterone and histamine levels in brain structures

Arginine vasopressin, injected systemically, significantly raised serum corticosterone levels measured 1 h later. Used in a dose of 5 μg/kg, AVP increased about four times this level; the latter dose was used in the present experiments (*Fig. 1*). Vasopressin did not substantially change the hypothalamic histamine levels, and markedly increased the histamine level in the hippocampus compared to saline-treated controls (*Fig. 2*).

The influence of α-FMH on the AVP-induced serum corticosterone and histamine levels in brain structures

The specific histamine biosynthesis inhibitor α-FMH (50 mg/kg ip), given 2 h before AVP, significantly diminished the AVP-elicited rise in the serum corticosterone level (by 36%) (*Fig. 3*). That diminution was correlated with a considerable decrease in the AVP-induced histamine content by α-FMH in the hypothalamus (by 68%), and a total reduction of its increase in the hippocampus (*Fig. 4*).

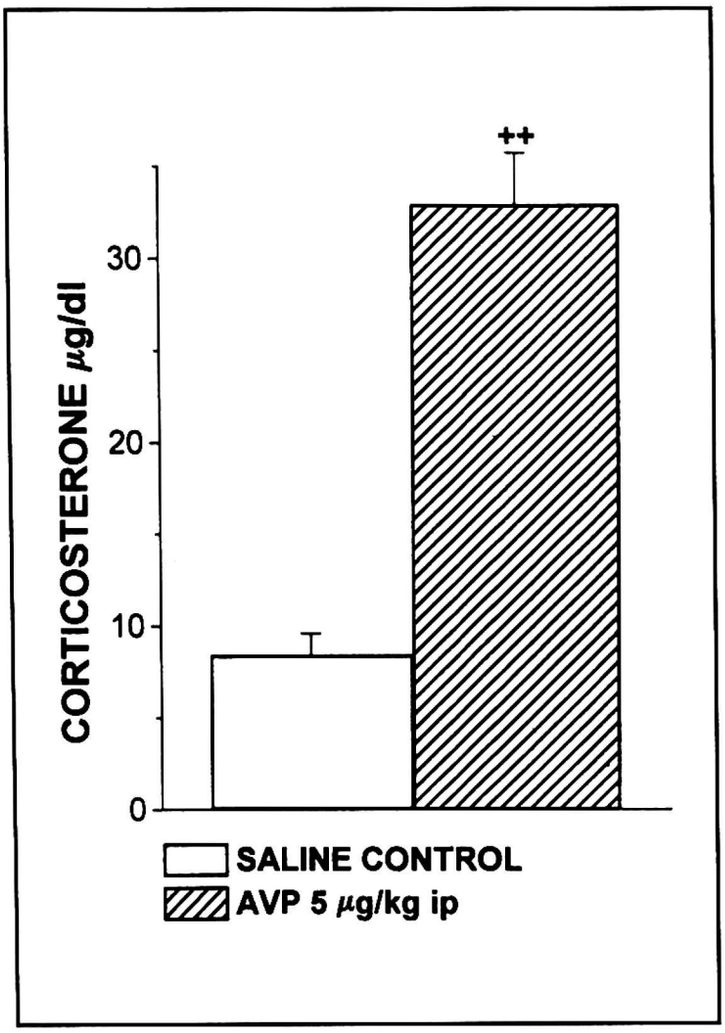

Fig. 1. Serum corticosterone concentration 1 h after ip vasopressin (AVP) injection. In *Fig. 1—8* values represent the mean ± SEM of 6 rats. $^{++}p < 0.01$ vs. saline controls.

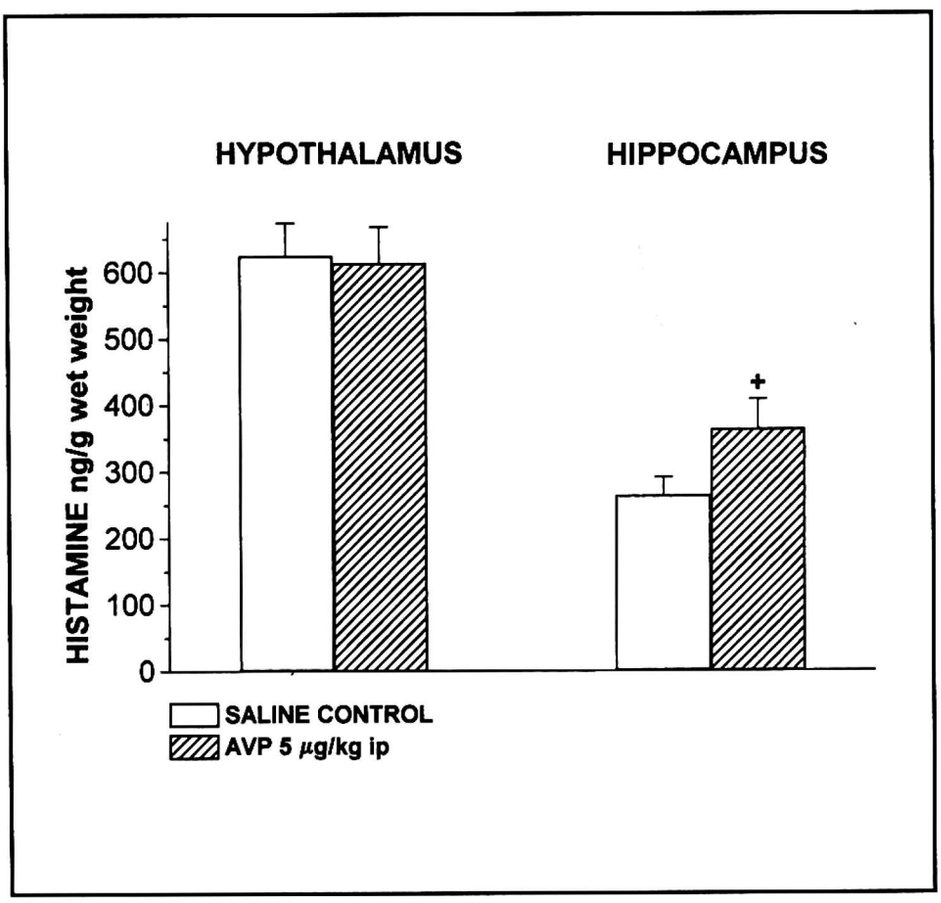

Fig. 2. Histamine content in the hypothalamus and hippocampus 1 h after ip vasopressin (AVP) injection. $^{+}p < 0.05$ vs. saline controls.

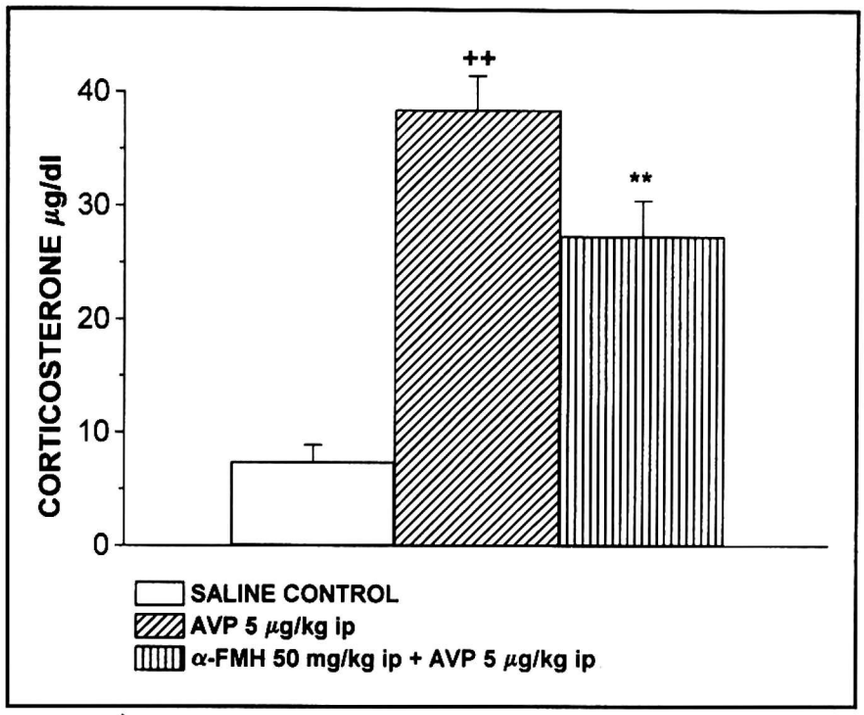

Fig. 3. Effect of α-fluoromethylhistidine (α-FMH) on the serum corticosterone concentration induced by vasopressin (AVP). α-fluoromethylhistidine was injected 2 h before AVP and 1 h later the rats were decapitated. $^{++}p < 0.01$ vs. saline controls and $^{**}p < 0.01$ vs. AVP-treated group.

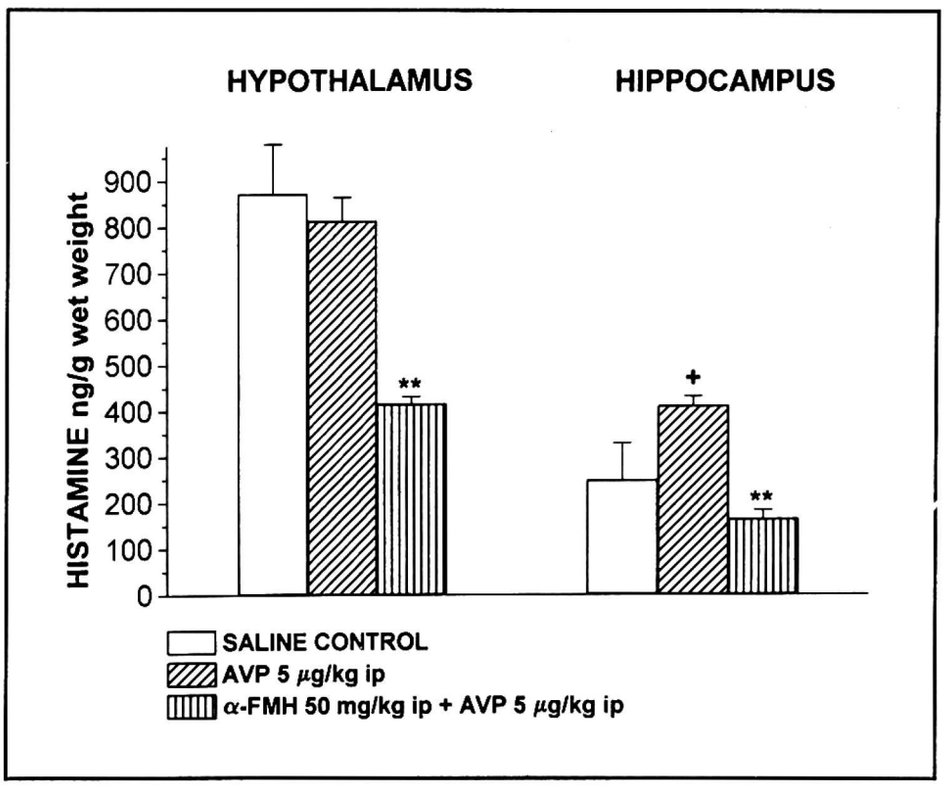

Fig. 4. Effect of α-fluoromethylhistidine on the histamine content in the hypothalamus and hippocampus induced by vasopressin. α-FMH was injected 2 h before AVP and 1 h later the rats were decapitated. $^{+}p < 0.05$ vs. saline controls and $^{**}p < 0.01$ vs. AVP-treated group.

Effect of histamine H_1- and H_2-receptor antagonists on the AVP-induced changes in the serum corticosterone and brain histamine levels

Systemic pretreatment with mepyramine (0.01 mg/kg ip), a histamine H_1-receptor antagonist, 15 min prior to AVP significantly diminished, by 45%, the vasopressin-induced corticosterone response. Ranitidine, (0.1 mg/kg ip), a histamine H_2-receptor antagonist, evoked an even stronger inhibition (62%) of the AVP-elicited corticosterone response (*Fig. 5*). The above findings suggest that both the histamine H_1- and H_2-receptors significantly mediate the

AVP-elicited HPA response. Pretreatment with either mepyramine or rantidine totally prevented the AVP-elicited increment in the hippocampal histamine content, but only mepyramine markedly decreased the hypothalamic histamine level (*Fig. 6*).

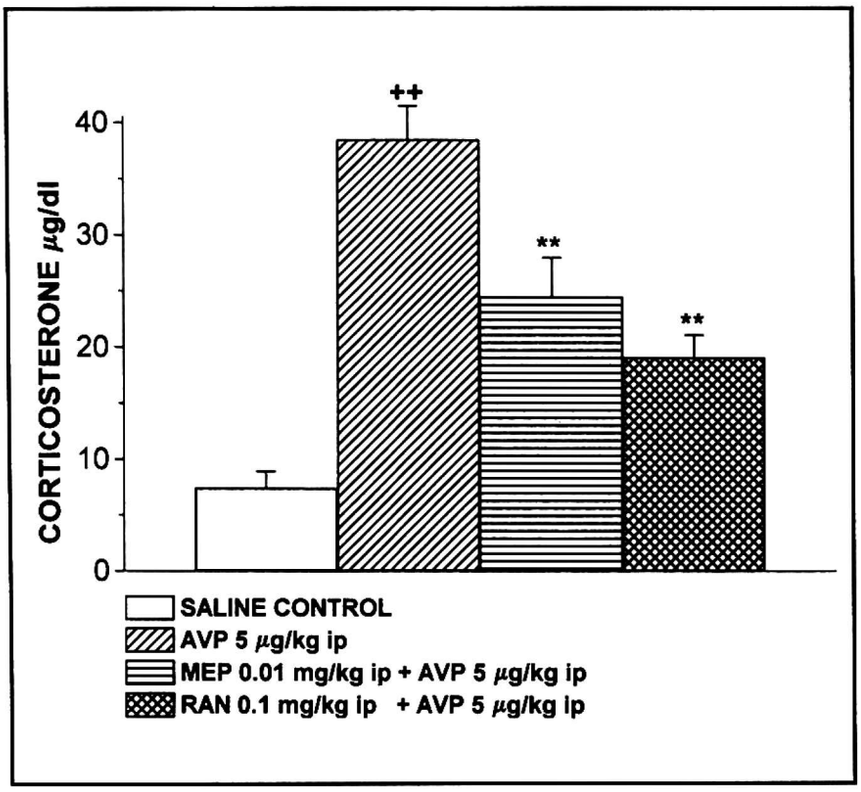

Fig. 5. Effect of mepyramine (MEP) and ranitidine (RAN) on the vasopressin-induced serum corticosterone concentration. MEP and RAN were injected 15 min before AVP and 1 h later the rats were decapitated. $^{++}p < 0.01$ vs. saline controls and $^{**}p < 0.01$ vs. AVP-treated group.

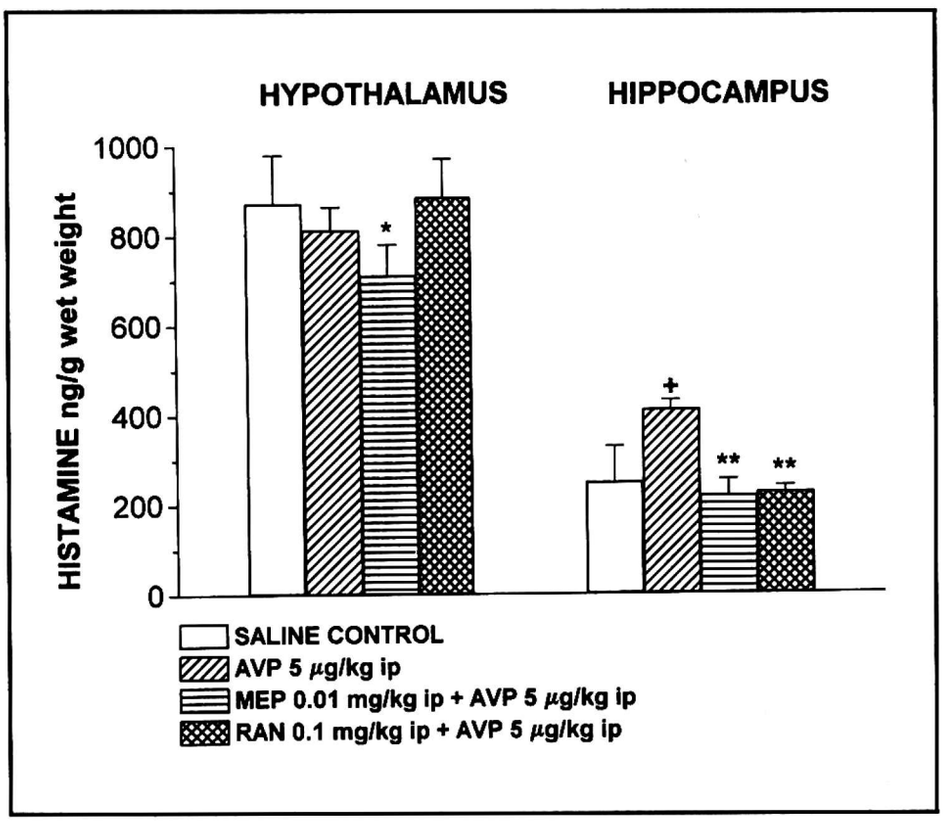

Fig. 6. Effect of mepyramine (MEP) and ranitidine (RAN) on the vasopressin-induced histamine content in the hypothalamus and hippocampus. MEP and RAN were injected 15 min before AVP and 1 h later the rats were decapitated. $^{+}p < 0.05$ vs. saline controls and $^{**}p < 0.01$ vs. AVP-treated group.

Effect of thioperamide on the vasopressin-induced changes in the serum corticosterone and brain histamine levels

Tioperamide (5 mg/kg ip), a specific histamine H_3-receptor antagonist, given 15 min prior to AVP significantly diminished (by 42%) the AVP-elicited increase in the serum corticosterone level (*Fig. 7*). However, this antagonist did not affect at all the AVP-induced hypothalamic and hippocampal histamine content (*Fig. 8*).

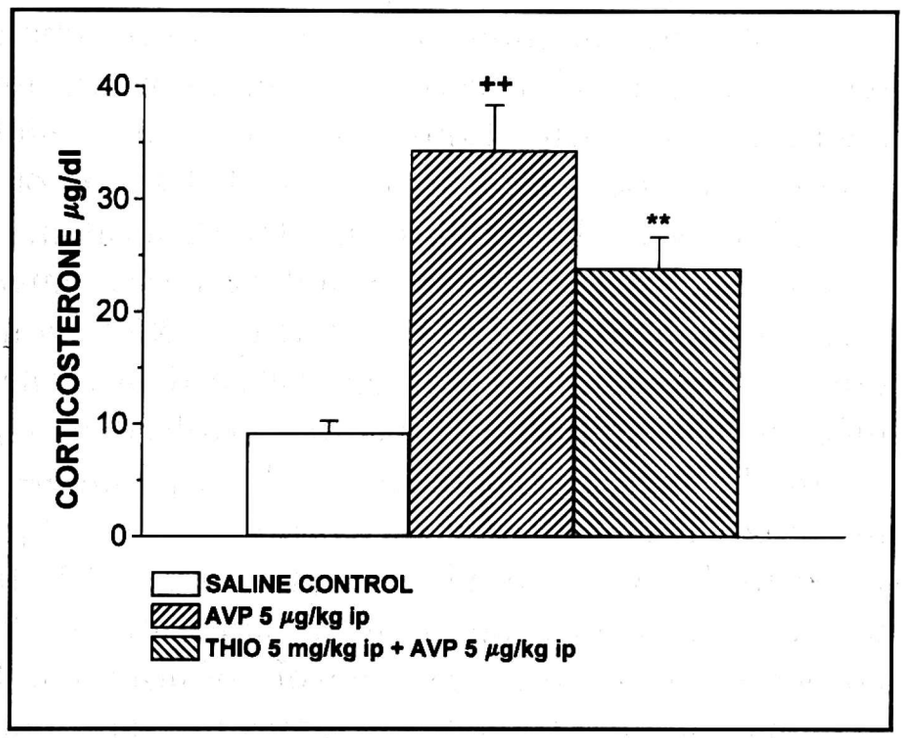

Fig. 7. Effect of thioperamide (THIO) on the vasopressin induced serum corticosterone level. THIO was injected 15 min before AVP and 1 h the rats were decapitated. $^{++}p < 0.01$ vs. saline controls and $^{**}p < 0.01$ vs. AVP-treated group.

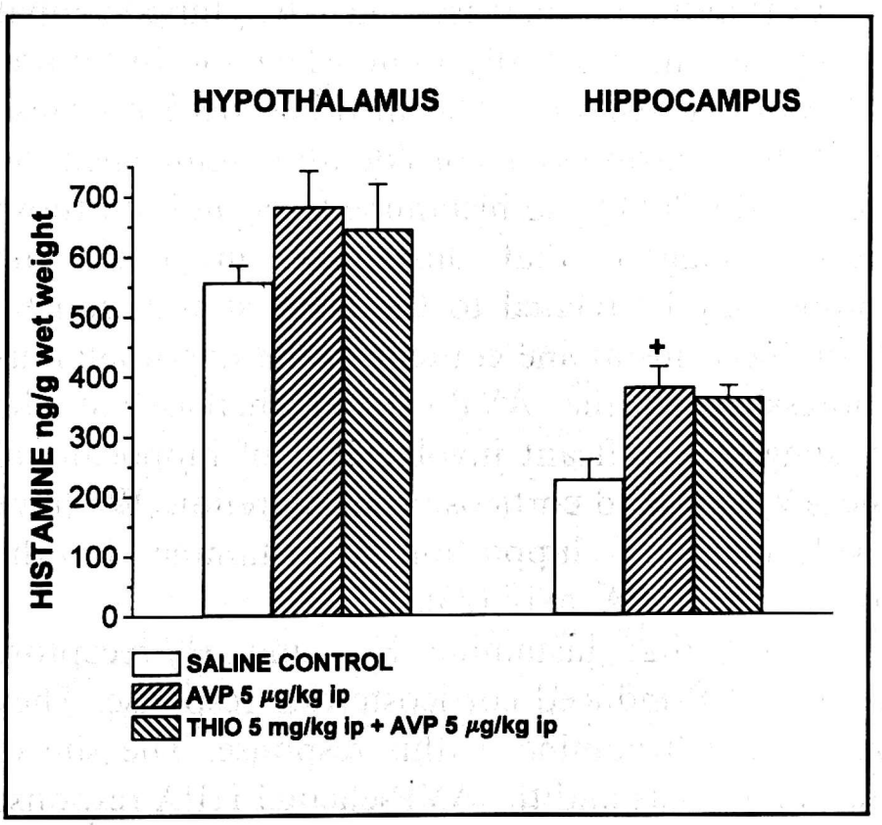

Fig. 8. Effect of thioperamide (THIO) on the vasopressin-induced histamine content in the hypothalamus and hippocampus. THIO was injected 15 min before AVP and 1 h later the rats were decapitated. $^{+}p < 0.04$ vs. saline controls.

DISCUSSION

The present study provides evidence for significant involvement of central histaminegic mechanisms in the pituitary-adrenocortical response to vasopressin. In agreement with our earlier data (8), AVP given systemically significantly increased the serum corticosterone concentration. Such an increase may be elicited by a direct activation of vasopressin receptors on anterior pituitary corticotrophs which leads to stimulation of the ACTH and corticosterone secretion, or by an indirect action *via* the endogenous CRH release (2, 21). After peripheral injection, vasopressin may also differently regulate its own expression in parvocellular neurons within the suprachiasmatic nucleus and the medial region of the hypothalamic PVN (22, 23). Moreover, also cerebrovascular permeability to circulating AVP at the blood-brain-barrier, and AVP uptake by circumventricular organs have been recently demonstrated (24). Therefore, AVP administered systemically may exert its stimulatory effect on the corticosterone secretion by activation of different suprapituitary structures involved in regulation of the HPA axis.

In the present experiment both mepyramine and ranitidine significantly impaired the AVP-elicited corticosterone response, by 45% and 62%, respectively. These results indicate that AVP stimulates the HPA axis *via* a histaminergic mechanism at the suprapituitary levels since anterior pituitary corticotrophs are practically devoid of histamine H_1- and H_2-receptors. After systemic administration, vasopressin did not substantially affect the hypothalamic histamine content, but markedly raised the histamine content in the hippocampus. Mepyramine and ranitidine, histamine H_1- and H_2-receptor antagonists, abolished the AVP-induced increases in the hippocampal histamine content, yet only mepyramine markedly diminished the histamine level in the hypothalamus of AVP-treated rats. The histidine decarboxylase inhibitor α-FMH, given ip 2 h before vasopressin, considerably diminished the AVP-elicited serum corticosterone (by 36%) and histamine levels in both those structures. These observations suggest that histamine in both the hypothalamus and hippocampus may be related to the HPA stimulation by AVP. A decrease in the AVP-induced histamine content in the hypothalamus by 68%, and a total suppression of the AVP-evoked increase in the hippocampal histamine may suggest significant involvement of hippocampal histamine in alterations in the AVP-induced corticosterone secretion. We have recently found similar involvement of hippocampal histamine in the clonidine-induced stimulation of the HPA axis (25).

These results evidently suggest that histamine H_1- and H_2-receptors mediate a significant part of the AVP-induced corticosterone response. They also point to a role of hippocampal histamine in this response. The site of interaction between histamine antagonists and the AVP-elicited HPA response

in the central nervous system is not known at present. Both the hypothalamus and hippocampus seem most likely to be involved in such intraction. The hypothalamic histaminergic system is a well-known as a stimulatory for the HPA axis (26, 27). Although a direct action of AVP on histamine receptors is not likely, this neurohormone may indirectly influence the central histaminergic system by interacting with an adrenergic mechanism, since AVP modulates the central adrenergic system (28).

The reduction of the AVP-induced response by pretreatment with α-FMH in the present experiment seems to be connected with significant diminution of the hypothalamic and hippocampal histamine content since α-FMH does not affect the catecholamine levels in these brain structure (29).

Our present results show that thioperamide, a histamine H_3-receptor antagonist (30), significantly diminishes the AVP-elicited corticosterone response but does not markedly affect the hypothalamic or hippocampal histamine levels. These findings suggest that thioperamide does not influence corticosterone secretion by accelerating histamine release from histaminergic neurons (31). In the present experiment direct adrenocortical inhibition at a non-H_3-intracellular site may rather be responsible for diminution of the AVP-induced corticosterone response (32).

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Author's address: J. Bugajski, Department of Physiology, Institute of Pharmacology, Polish Academy of Sciences, Smętna 12, 31-343 Cracow, Poland.