Hydruric Response in Wistar and Vasopressin-Deficient Brattleboro Rats to Water Load under Conditions of Increased Brain Serotonin Level

L. N. Ivanova, L. N. Kochkaeva, N. N. Melidi

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 141, No. 2, pp. 137-139, February, 2006
Original article submitted March 15, 2005

The hydruric response to water load in Wistar rats and homozygous Brattleboro rats with a hereditary defect in the synthesis of vasopressin was studied under conditions of increased brain serotonin level. Serotonin prevented the reduction in reabsorption of osmotically free water in normal rats, but had no effect in vasopressin-deficient Brattleboro rats. Our results suggest that serotonin stimulates vasopressin secretion and interacts with the vasopressinergic system during the realization of osmotic regulation.

Key Words: *serotonin; vasopressin; hydration; renal function; Brattleboro rats*

The neurohypophyseal hormone vasopressin regulates water excretion in the kidneys and plays a major role in the multicomponent system that maintains water balance in mammals and humans. Physiological activity of vasopressin is not limited to its antidiuretic effect. Vasopressin is one of the major vasoconstrictors. This compound modulates ACTH secretion, affects training and memory consolidation, possesses mitotic activity, stimulates immunoglobulin syntheses in lymphocytes, and promotes proliferation and differentiation of T lymphocytes [10,14]. Due to polyfunctional activity of vasopressin, the biosynthesis and secretion of this substance are regulated by various neurochemical systems of the brain [5,13]. Published data show that secretion of vasopressin depends on serotoninergic influences [8]. Vasopressin, in turn, activates serotonin biosynthesis in the midbrain and anterior hypothalamus. The reaction of the cerebral serotoninergic system to dehydration is modified in Brattleboro rats with genetically determined absence of vasopressin [2,12]. These data indicate that water balance is regulated by 2 neurochemical systems. However, functional significance of the interaction between the serotoninergic and vasopressinergic systems in the realization of osmoregulatory reactions remains unknown. Here we studied the hydruric response to water load in Wistar and vasopressin-deficient Brattleboro rats to increased brain serotonin level.

MATERIALS AND METHODS

Experiments were performed on adult conscious Wistar rats (200-250 g) and homozygous vasopressin-deficient Brattleboro rats obtained from the vivarium of the Institute of Cytology and Genetics. The animals were kept in a vivarium under standard conditions. The rats were placed in individual metabolic cages, which allowed us to collect spontaneous urine samples and determine renal function. Basal urine samples were collected from control rats over the 1st hour of the study. Water (5% body weight) was administered intragastrically through a polyethylene probe. The animals simultaneously received intraperitoneal injection of 600 μl physiological saline. Urine samples were collected over

Laboratory of Physiological Genetics, Institute of Cytology and Genetics, Siberian Division of the Russian Academy of Sciences, Novosibirsk. ***Address for correspondence:*** ludiv@bionet.nsc.ru. L. N. Ivanova.

0007-4888/06/1412-0178

the next 4 h. The experimental design did not differ for animals of various groups. Water load in experimental animals was accompanied by intraperitoneal injection of a direct serotonin precursor 5-hydroxytryptophan (50 mg/kg, Calbiochem) to increase brain serotonin level. 5-Hydroxytryptophan rapidly crosses the blood-brain barrier and induces an immediate increase in serotonin concentration, which is spontaneously released into the cerebrospinal fluid [3]. The time to peak response and effectiveness of water excretion (% of the initial water volume) were estimated in each animal. Blood samples were collected after decapitation. Renal function and response to treatment were evaluated by diuresis (V), glomerular filtration rate (GFR), and reabsorption of osmotically free water ($T^f_{H_2O}$). The glomerular filtration rate was estimated in the endogenous creatinine clearance test. Osmolality of the blood and urine samples was measured by the cryoscopic method on a MT-2 milliosmometer. Parameters of renal function were calculated routinely [1]. Changes in plasma vasopressin level were estimated by reabsorption of osmotically free water.

The results were analyzed by Student's *t* test.

RESULTS

Water load induced a typical hydruric response in Wistar rats, which was determined by decreased plasma vasopressin level: diuresis increased due to a sharp decrease in reabsorption of osmotically free water and acceleration of glomerular filtration (Table 1). The hydruric response was most pronounced 1-1.5 h after intragastric administration of water. The administered volume of fluid was excreted over 4 h. The hydruric response was not observed after administration of 5-hydroxytryptophan, *i.e.* under conditions of increased brain serotonin level. Renal function little changed under these conditions. Reabsorption of osmotically free water remained positive. Diuresis increased only by the end of the study. The animals excreted only 40% of administered water.

Water load had little effect on reabsorption of osmotically free water in control Brattleboro rats. $T^f_{H_2O}$ tended to decrease in these animals. However, these differences were statistically insignificant due to large dispersion of experimental data. The glomerular filtration rate increased. The volume of fluid excreted over 4 h surpassed the volume of water load in Brattleboro rats. It was related to intensive diuresis throughout the study and absence of endogenous vasopressin (negative reabsorption of osmotically free water). As differentiated from Wistar rats, 5-hydroxytryptophan-induced increase in brain serotonin level in vasopressin-deficient Brattleboro rats had no effect on renal function. Reabsorption of osmotically free water remained negative, which is characteristic of the basal state and water load.

There are ambiguous data on the effect of brain serotonin on the synthesis and secretion of vasopressin. Some authors showed that serotonin inhi-

TABLE 1. Effect of 5-Hydroxytryptophan on Diuresis and Renal Function in Wistar and Brattleboro Rats (*M*±*m*)

Parameter	Water excretion over 4 h after water load, %	V, ml/h per 100 g wet tissue weight		GRF, ml/h per 100 g wet tissue weight		$T^f_{H_2O}$, ml/h per 100 g wet tissue weight	
		basal	max	basal	max	basal	max
Wistar							
Water load and physiological saline	102±9 (8)	0.520±0.027 (8)	5.17±0.53*** (8)	14.6±2.6 (8)	29.4±5.4** (8)	0.48±0.10 (8)	-4.12±0.69*** (8)
Water loading and 5-hydroxytryptophan	40.0±13.6^{++} (7)	0.54±0.32 (7)	0.18±0.09^{++} (7)	17.1±4.8 (7)	10.7±3.5^{+} (7)	0.35±0.15 (7)	0.36±0.17^{+++} (7)
Brattleboro							
Water load and physiological saline	140±21 (8)	1.2±0.3 (8)	3.9±1.9 (8)	13.9±1.6 (8)	32.3±9.6* (8)	-0.60±0.26 (8)	-2.2±1.3 (8)
Water loading and 5-hydroxytryptophan	132±19 (8)	1.7±0.3 (8)	3.1±1.9 (8)	14.9±4.9 (8)	37.4±8.8* (8)	-0.83±0.31 (8)	-2.2±1.2 (8)

Note. Number of animals is shown in brackets. *p<0.05, **p<0.01, and ***p<0.001 compared to the basal level; ^{+}p<0.02, ^{++}p<0.002, and ^{+++}p<0.001 compared to water load and physiological saline.

bits [4,6], while others reported that this compound has no effect on vasopressin secretion *in vivo* [15] and *in vitro* [7]. Our results show that serotonin modulates typical hydruric response to water load and prevents inhibition of vasopressin secretion in Wistar rats. The inhibition of reabsorption of osmotically free water in vasopressin-deficient Brattleboro rats was observed during water load, but did not occur under conditions of increased brain serotonin level. 5-Hydroxytryptophan was ineffective in these animals. These data suggest that serotonin contributes to the release of vasopressin into the circulation. Our findings are consistent with published data that central treatment with serotonin and increase in the concentration of endogenous serotonin prevent reuptake of the transmitter and stimulate secretion of vasopressin into the peripheral blood [9,11].

The serotoninergic and vasopressinergic systems play a complex role in osmotic regulation. The discrepancy in the data on serotonin-induced stimulation of vasopressin secretion is partly due to various experimental conditions and realization of the reaction via various types of serotonin receptors. Activation of vasopressin secretion with serotonin under conditions of water load is probably mediated by 5-HT_{2C}, 5-HT_4, and 5-HT_7 receptors. Previous studies of vasopressin secretion after treatment with agonists and antagonists of various types of serotonin receptors showed that these receptors mediate activation of vasopressin secretion by intracerebroventricular drugs [8].

Our results suggest that the serotoninergic and vasopressinergic systems produce a complex regulatory effect on water balance. The osmoregulatory response is probably influenced by the increase in brain serotonin level.

This work was supported by the Russian Foundation for Basic Research (grant No. 04-04-49075), and Foundation of the President of Russian Federation for Support of Leading Scientific Schools (grant No. NSh-1515.2003.4).

REFERENCES

1. Yu. V. Natochin *Renal Physiology. Formulas and Calculations* [in Russian], Leningrad (1974).
2. N. K. Popova, L. N. Ivanova, T. G. Amstislavskaya, *et al., Ros. Fiziol. Zh*., **86**, No. 2, 140-147 (2000).
3. N. K. Popova, E. V. Naumenko, and V. G. Kolpakov, *Serotonin and Behavior* [in Russian], Novosibirsk (1978).
4. M. Altemus, G. Cizza, and P. W. Gold, *Brain Res*., **593**, No. 2, 311-313 (1992).
5. N. Falk, *Prog. Neurobiol*., **36**, 465-484 (1991).
6. C. F. Ferris, *Ann. NY Acad. Sci*., **794**, 98-103 (1996).
7. E. W. Hillhouse and N. G. N. Milton, *J. Endocrinol*., **122**, 713-718 (1989).
8. H. Jorgensen, M. Riis, U. Knigge, *et al., J. Neuroendocrinol*., **15**, 242-249 (2003).
9. R. Montes and A. K. Johnson, *Am. J. Physiol*., **259**, R1267-R1273 (1990).
10. A. Pereira, M. Del Valle Onorato, and C. Sanz, *Br. J. Haemat.*, **120**, No. 5, 814-820 (2003).
11. P. E. Pergola, A. F. Sved, J. L. Voogt, *et al., J. Neuroendocrinol*., **57**, 550-558 (1993).
12. N. K. Popova, K. S. Naumenko, L. N. Maslova *et al., Pflugers Arch. Eur. J. Physiol*., **444**, 372-377 (2002).
13. S. Reichlin, *Textbook of Endocrinology*, Eds. J. D. Wilson *et al.,* Philadelphia (1998), pp. 165-248.
14. R. M. A. Richardson, *Curr. Nephrol*., **15**, 149-205 (1992).
15. L. D. Van de Kar, A. D. Levy, Q. Li, *et al., Pharmacol. Biochem. Behav*., **60**, No. 3, 677-683 (1998).