*Acta Medica Okayama*

*Volume* 50, *Issue* 4 1996 *Article* 5

AUGUST 1996

# Platelet Taurine Concentration and Uptake in the Brattleboro Diabetes Insipidus Rat

Marja-Leena Nieminen* Leena Tuomisto†

Erkka Solatunturi‡

*University of Helsinki,

†University of Kuopio,

‡University of Kelsinki,

Copyright ©1999 OKAYAMA UNIVERSITY MEDICAL SCHOOL. All rights reserved.

# Platelet Taurine Concentration and Uptake in the Brattleboro Diabetes Insipidus Rat*

Marja-Leena Nieminen, Leena Tuomisto, and Erkka Solatunturi

## Abstract

Since platelets accumulate taurine, they provide a model for studying the taurine transport in anisosmotic disorders. Thus, in this work we studied the taurine concentration and uptake in the platelets of Brattleboro rats, homozygous (DI) and heterozygous (HZ) for hereditary hypothalamic diabetes insipidus, and Long Evans (LE) normal rats after free water intake and after dehydration induced by water deprivation for 24 h. The decreased ability of the DI rats to concentrate urine led to plasma hypernatremia and hyperosmolality despite excessive drinking. Water deprivation in the DI rats induced drastic dehydration with exacerbated hypernatremia and hyperosmolality. Plasma hypernatremia and hyperosmolality resulted in a significant elevation of the taurine concentration and uptake by platelets of the DI rats. Kinetic assays showed that plasma hypernatremia and hyperosmolality did not alter the affinity of taurine to platelet membrane carrier, as expressed by Km, but caused a profound increase in the maximal transport capacity, Vmax. After free water intake the Vmax of the DI rats was about two times higher than that in the HZ and LE rats and after water deprivation it was about three times higher. Water deprivation doubled the Vmax of the DI rats without changing the Km.

**KEYWORDS:** Brattleboro diabetes insipidus rat, platelet, taurine concentrationm taurine uptake, osmoregulation

---

*PMID: 8874582 [PubMed - indexed for MEDLINE]
Copyright (C) OKAYAMA UNIVERSITY MEDICAL SCHOOL

Acta Med Okayama 1996; 50(4): 203 210

# Platelet Taurine Concentration and Uptake in the Brattleboro Diabetes Insipidus Rat

Marja-Leena Nieminen*, Leena Tuomisto[a] and Erkka Solatunturi

Institute of Biomedicine, Department of Pharmacology and Toxicology, University of Helsinki, Helsinki 00014, Finland and [a]Department of Pharmacology and Toxicology, University of Kuopio, Kuopio 70211, Finland

Since platelets accumulate taurine, they provide a model for studying the taurine transport in anisosmotic disorders. Thus, in this work we studied the taurine concentration and uptake in the platelets of Brattleboro rats, homozygous (DI) and heterozygous (HZ) for hereditary hypothalamic diabetes insipidus, and Long Evans (LE) normal rats after free water intake and after dehydration induced by water deprivation for 24 h. The decreased ability of the DI rats to concentrate urine led to plasma hypernatremia and hyperosmolality despite excessive drinking. Water deprivation in the DI rats induced drastic dehydration with exacerbated hypernatremia and hyperosmolality. Plasma hypernatremia and hyperosmolality resulted in a significant elevation of the taurine concentration and uptake by platelets of the DI rats. Kinetic assays showed that plasma hypernatremia and hyperosmolality did not alter the affinity of taurine to platelet membrane carrier, as expressed by $K_m$, but caused a profound increase in the maximal transport capacity, $V_{max}$. After free water intake the $V_{max}$ of the DI rats was about two times higher than that in the HZ and LE rats and after water deprivation it was about three times higher. Water deprivation doubled the $V_{max}$ of the DI rats without changing the $K_m$.

**Key words:** Brattleboro diabetes insipidus rat, platelet, taurine concentration, taurine uptake, osmoregulation

It is well-documented that taurine serves as an osmoregulator in marine invertebrates, teleosts and amphibians. During adaptation to increased or decreased environmental salinity, taurine levels in the tissues of these marine animals increase or decrease, respectively, to limit the loss or gain of water in tissues and to maintain osmotic equilibrium (1, 2).

There is also considerable evidence that supports a role for taurine in the mechanisms of cell volume regulation in mammals. The response of mammalian cells to hypoosmolality by an adaptive decrease of the taurine content has been demonstrated in many preparations, ranging from cultured cells to intact tissues (3–10). It has also been shown that an adaptive response of mammalian cells to hyperosmolality involves an increase in the concentration of cellular taurine. Thurston *et al*, (11, 12) were the first to show that in weanling mice chronic hypernatremic dehydration increased the taurine concentration in the brain and heart. More recently, we have demonstrated increased taurine levels in the brain, skeletal muscle and platelets of the chronically hypernatremic Brattleboro rat with hereditary hypothalamic diabetes insipidus (13).

The mechanisms responsible for the adaptive increase of cellular taurine with plasma hypernatremia are not known. The increase may result from stimulation of the taurine uptake system. Since platelets accumulate taurine, they may provide a convenient model for studies on taurine transport in cells. In order to obtain information about the mechanisms responsible for increased cellular taurine associated with plasma hypernatremia, we compared the taurine concentration and the kinetics of taurine uptake in platelets of the Brattleboro rat after free water intake and after dehydration induced by water deprivation.

## Materials and Methods

***Animals.*** Male rats of the Brattleboro strain, homozygous (DI) and heterozygous (HZ) for hypoth-

* To whom correspondence should be addressed.

Acta Medica Okayama, Vol. 50 [1996], Iss. 4, Art. 5



alamic diabetes insipidus, and Long Evans (LE) hooded rats, genetic controls for DI and HZ rats, were used. The rats were bred at the Department of Pharmacology and Toxicology, University of Helsinki, and at the National Laboratory Animal Center, University of Kuopio, Finland. At the beginning of the experiment the rats were 6 months old and the experiment took approximately 6 weeks to complete.

***Experimental procedure.*** Before the experiment, the rats were allowed to adapt to metabolic cages for 1 week with free access to tap water and food (Ewos R3, Ewos Sverige AB, Södertälje, Sweden). Within each genotype, the rats were randomly assigned either to the water-deprived group or to the water-control group. In the water-deprived group, dehydration was induced by complete water deprivation for 24 h while food was available *ad libitum*. In the water-control group, the rats were given free access to water and food. Water intake was measured for 24 h. The rats were weighed at the beginning and at the end of the 24-h experimental period. Urine was collected for 24 h and its volume and osmolality were recorded.

***Preparation of platelet-rich plasma.*** After the 24-h experimental period, the rats were killed under ether anesthesia by draining the blood from the carotid artery into a polypropylene tube containing 1/10 volume 1.5 % dinatrium edetate as an anticoagulant. The platelet-rich plasma was obtained by centrifuging the blood at $150 \times g$ for 20 min at room temperature, and by pipetting the upper plasma layer into another polypropylene tube. When necessary, the plasmas of two rats were pooled.

***Taurine concentration in platelets.*** The taurine concentration in platelets, obtained by centrifugation of the platelet-rich plasma at $12,000 \times g$ for 10 min, was estimated as a fluorescamine derivative after purification on ion-exchange column (14). The taurine concentration is expressed as $\mu$mol/$10^{12}$ platelets. The platelet counts, platelets/$\mu$l of the platelet-rich plasma, were done under a phase contrast microscope using a Bürker chamber (16).

***Taurine uptake by platelets.*** Taurine uptake was studied in samples containing 1/3 volume plasma and 200,000 platelets/$\mu$l. For this, the platelet-rich plasma was first diluted with platelet-free plasma, obtained by further centrifuging the rest of the blood sample at high speed, to adjust the platelet count to 600,000 platelets/$\mu$l. After that, plasma samples were diluted with 2 volumes of $Ca^{++}$-$Mg^{++}$-free Krebs-Ringer solution to give the final platelet count of 200,000 platelets/$\mu$l.

Taurine uptake by platelets was studied as a function of incubation time and at various substrate concentrations to calculate kinetic parameters.

For the time course of taurine uptake, duplicate samples (1.0 ml) were preincubated in an atmosphere of 95 % $O_2$ and 5 % $CO_2$ for 10 min at 37 °C in a shaking water bath. After preincubation, 5 $\mu$M [$^3$H] taurine (specific activity 3.7 GBg/mol, New England Nuclear, Boston, Mass., USA) was added and the influence of incubation time on taurine uptake by platelets was measured after 5, 10, 20, 30, 60, and 120 min. The uptake reaction was terminated by placing the sample tubes in an ice bath. The radioactivity of the platelets was assayed by liquid scintillation counting (1219 Rack Beta Liquid Scintillation Counter, LKB Wallac, Turku, Finland) after washing and ultrasonic disruption of the cells. To determine whether taurine uptake by platelets is temperature dependent, the above experiment was repeated at 0 °C in an ice bath.

For kinetic parameters, the determination of taurine uptake was performed according to the method of Arora and Meltzer for 5-hydroxytryptamine (17, 18). Active uptake was determined by the difference in the uptake values between one set of sample tubes incubated after prewarming for a desired time at 37 °C (total uptake) and another set placed in an ice bath to 0 °C (passive diffusion) immediately after adding [$^3$H] taurine at six different concentrations ranging between 1 $\mu$M and 40 $\mu$M. The maximal velocity ($V_{max}$) and the Michaelis constant ($K_m$) were calculated by means of a computer program using the Lineweaver-Burk method (19, 20). $V_{max}$ is expressed as nmol/$10^{12}$ platelets/10 min. $K_m$ is expressed in micromoles.

***Measurement of osmolality and sodium concentration.*** The osmolality and sodium concentration of the platelet-free plasma, obtained by centrifuging the platelet-rich plasma at $12,000 \times g$ for 10 min, were recorded. The sodium was measured by flame photometry (IL Model 243 Flame Fotometer, Instrumentation Laboratory, USA) and the osmolality of plasma and urine with a vapour pressure osmometer (Model 5,100 C, Wescor Inc., Utah, USA).

***Statistics.*** Data are expressed as means ± standard erros (S. E.). Statistical significance was determined using the analysis of variance (ANOVA) for multiple comparisons and Student's *t*-test for paired comparisons.

Statistical significance was considered at $P < 0.05$.

## Results

Rats of the Brattleboro strain can be divided into three genotypes: Brattleboro homozygotes (DI) completely lacking vasopressin, Brattleboro heterozygotes (HZ) with vasopressin synthesis of about 50–60 % that in normal rats, and normal rats of the Long Evans (LE) hooded strain from which Brattleboro rats are derived (23). The major features of fluid balance in Brattleboro rats after water intake and after water deprivation of 24 h are shown in Figs. 1 and 2 and in Table 1.

### *Fluid Balance after Free Water Intake*

The mean daily urine volume of the DI rats, reflecting the severity of diabetes insipidus, was about 53 % of the body weight, and the urine showed a mean osmolality of 180 mOsm/kg. Conversely, in the control LE rats, the daily urine flow averaged 2 % of the body weight, and their urine osmolality was about 2,100 mOsm/kg. In the HZ rats, the values for fluid balance fell between those for the control LE rats and the DI rats, urine volume being 4 % of the body weight and urine osmolality about 1,100 mOsm/kg. After free water intake for 24 h the changes in body weight were about ± 2 % in all three rat genotypes (Table 1). Although the daily water intake exceeded the renal water loss, the plasma sodium concentration, 166 ± 1 mmol/l (mean ± S. E.) and plasma osmolality, 319 ± 2 mOsm/kg (mean ± S. E.) of the DI rats were significantly higher than those of the HZ rats (159 ± 1 mmol/l and 305 ± 4 mOsm/kg) and those of

**Table I** Changes of body weight in Brattleboro homozygotes (DI), Brattleboro heterozygotes (HZ) and Long Evans (LE) normal rats after free water intake for 24 h [water-control group (I)] and after water deprivation for 24 h [water-deprived group (II)].

| | LE | HZ | DI |
|---|---|---|---|
| Free water intake (I) | | | |
| Initial weight (g) | 362 ± 12 | 480 ± 27 | 379 ± 7 |
| Final weight (g) | 355 ± 13 | 488 ± 20 | 371 ± 9 |
| | (n = 7) | (n = 5) | (n = 7) |
| Change (%) | − 2 ± 1 | + 2 ± 2 | − 2 ± 2 |
| Water deprivation (II) | | | |
| Initial weight (g) | 388 ± 6 | 457 ± 9 | 393 ± 15 |
| Final weight (g) | 365 ± 4 | 429 ± 7 | 344 ± 14 |
| | (n = 7) | (n = 7) | (n = 10) |
| Change (%) | − 6 ± 1[a] | − 6 ± 1[a] | − 12 ± 1[b] |

Numbers of animals are shown in parenthesis. Means ± S. E. are given.
Significance of differences: [a] I *vs.* II, $P < 0.05$; [b] I *vs.* II and DI *vs.* LE, HZ $P < 0.001$

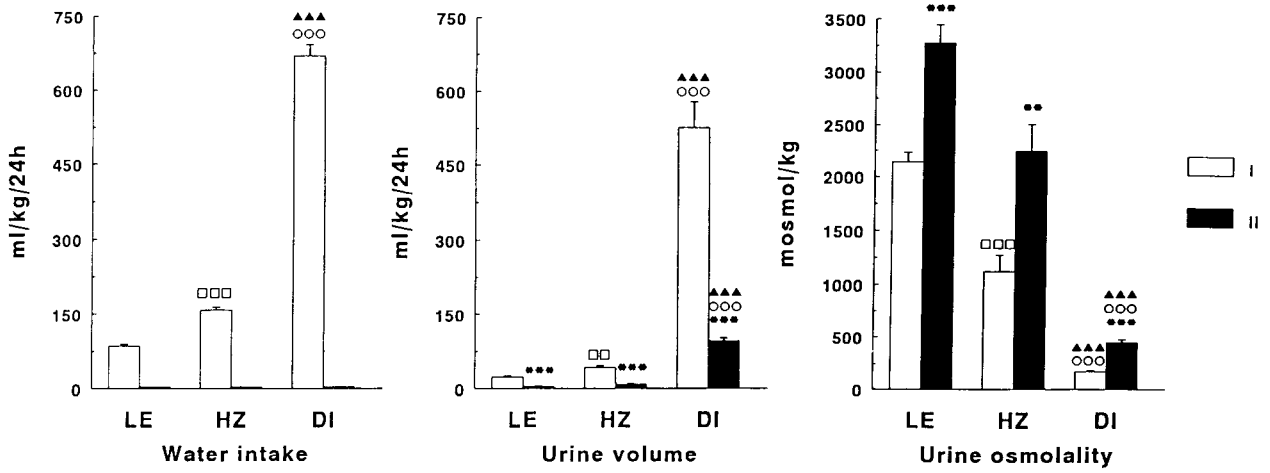


**Fig. 1** Water intake, urine volume and urine osmolality of Brattleboro homozygotes (DI), Brattleboro heterozygotes (HZ) and Long Evans (LE) normal rats after free water intake for 24 h (water-control group (I), open bars) and after water deprivation for 24 h (water-deprived group (II), filled bars). Means ± S. E. from 5 to 10 animals are given. The symbols for the significance of differences: I *vs.* II: $*P < 0.05$, $**P < 0.01$, $***P < 0.001$; DI *vs.* LE: ○ $P < 0.05$, ○○ $P < 0.01$, ○○○ $P < 0.001$; DI *vs.* HZ: ▲ $P < 0.05$, ▲▲ $P < 0.01$, ▲▲▲ $P < 0.001$; HZ *vs.* LE: □ $P < 0.05$, □□ $P < 0.01$, □□□ $P < 0.001$.

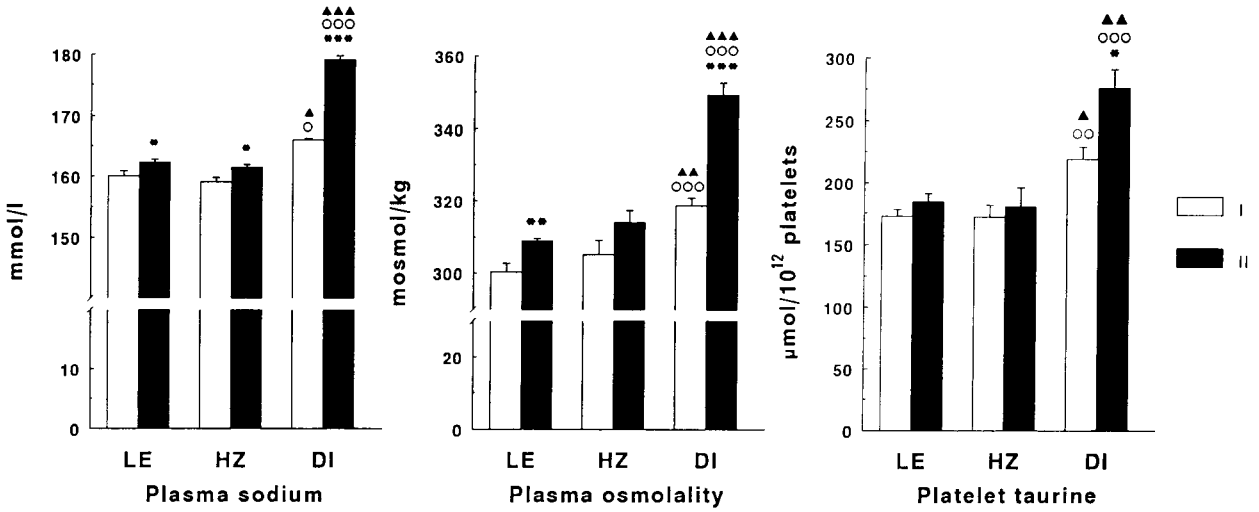


**Fig. 2** Plasma sodium concentration, plasma osmolality and platelet taurine of Brattleboro homozygotes (DI), Brattleboro heterozygotes (HZ) and Long Evans (LE) normal rats after free water intake for 24 h (water-control group (I), open bars) and after water deprivation for 24 h (water-deprived group (II), filled bars). Means ± S. E. from 5 to 10 animals (in platelet taurine from 4 to 6 animals) are given. Two-way ANOVA for sodium concentration and osmolality: group effect, $P < 0.001$, genotype effect $P < 0.001$ and group × genotype interaction, $P < 0.001$. The symbols for the significance of differences as in Fig. 1.

the LE rats (160 ± 1 mmol/l and 300 ± 2 mOsm/kg).

### *Fluid Balance after Water Deprivation*

When deprived of drinking water, the inability of the DI rats to concentrate urine results very quickly in severe dehydration. Over a 24-h water deprivation, the DI rats lost 12 % of their boby weight, and their plasma sodium concentration, 179 ± 1 mmol/l was about 8 % higher than that of the DI rats during free water intake. In addition, the plasma osmolality (349 ± 3 mOsm/kg) increased by nearly 10 % over the same period, indicating a marked concentration of body fluids during water deprivation. Over a 24-h period of water deprivation, the LE rats and also the HZ rats with a partial defect in vasopressin synthesis showed a high urine osmolality, up to 3,300 for the LE rats and 2,200 mOsm/kg for the HZ rats. The loss of body weight was about 6 % in both the HZ and the LE rats during water deprivation. In both genotypes, the plasma sodium (161 ± 1 mmol/l for HZ and 162 ± 1 mmol/l for LE) increased by only a little over 1 % and the plasma osmolality (314 ± 3 mOsm/kg for HZ and 309 ± 1 mOsm/kg for LE) increased by about 3 % as compared to the same genotype after free water intake.

### *Taurine Concentration in Platelets*

If taurine is functioning as an active osmolyte, it should increase in tissues under hyperosmotic conditions. Such a response can be seen in Fig. 2 where the elevation of plasma sodium and osmolality has induced an adaptive increase in platelet taurine. Even in free water intake, the taurine concentration in platelets of the DI rats is nearly 30 % higher than that of the HZ and LE rats due to about 5 % higher plasma sodium and osmolality. In response to dehydration induced by water deprivation of 24 h, platelet taurine of the DI rats further increased, the concentration being almost 50 % higher than that of the HZ and LE rats, and this is associated with more than 10 % higher plasma sodium and osmolality.

### *Taurine Uptake*

***Time course of taurine uptake.*** Fig. 3 shows the time course of taurine uptake by platelets of the Brattleboro rat at 37 °C. The uptake of taurine by platelets followed a linear course at least for the first 10 min, reached a maximum at 30 to 60 min, and then began to decline. Water deprivation increased the taurine uptake in all three genotypes of Brattleboro rat. The platelets of the DI rats accumulated more taurine than those of the HZ and LE rats. After water deprivation, the taurine uptake of the DI rats was twofold that of the LE and HZ rats throughout the incubation period of 120 min. Taurine uptake at 0 °C was negligible.

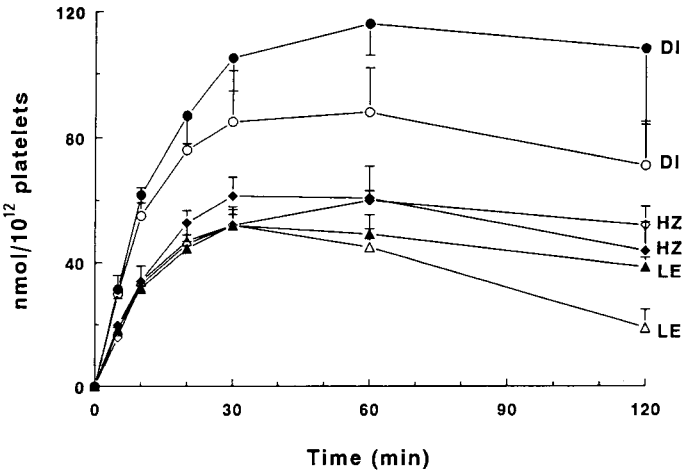


**Fig. 3** Time course of taurine uptake by platelets of Brattleboro homozygotes (DI), Brattleboro heterozygotes (HZ) and Long Evans (LE) normal rats after free water intake for 24 h (open symbols) and after water deprivation for 24 h (filled symbols). Platelets were incubated with 5 $\mu$M [$^3$H] taurine at 37 °C. Taurine uptake at 0 °C was under 10 nmol in all the three genotypes throughout the incubation period of 120 min. Each point represents the mean value ± S.E. of 3–5 experiments.

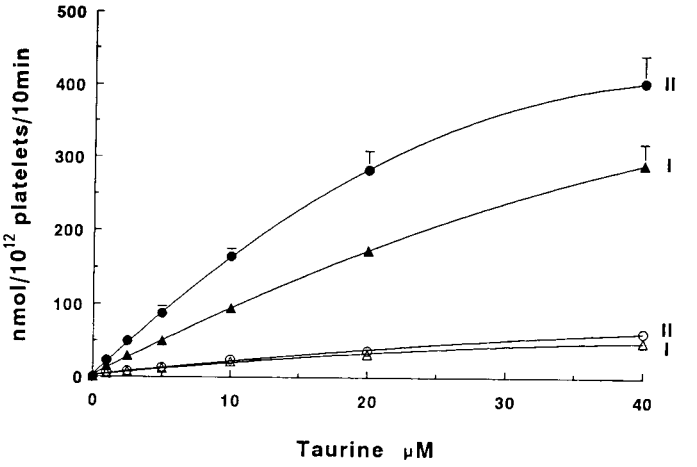


**Fig. 4** Taurine uptake by platelets of Brattleboro homozygotes (DI) after free water intake for 24 h (I) and after water deprivation for 24 h (II). Platelets were incubated at 37 °C (filled symbols) and at 0 °C (open symbols) with various concentrations of [$^3$H] taurine. Each point represents the mean value ± S.E. of 7–10 experiments.

***Kinetics of taurine uptake.*** To determine the kinetic constants of taurine uptake by platelets of the Brattleboro rat, transport studies were performed over a taurine concentration range of 1–40 $\mu$M. Because linear uptake continued at least for the first 10 min, the rate of influx within this period was taken as the initial rate of uptake. The active uptake was calculated by substracting the uptake at 0 °C, used to illustrate passive diffusion, from the total uptake at 37 °C.

As shown in Fig. 4, water intake or deprivation had no effect on taurine uptake at 0 °C, illustrating passive diffusion. There was also no difference between the three genotypes of the Brattleboro rat in taurine uptake at 0 °C.

Taurine uptake by platelets showed a saturating tendency (Fig. 5A). Within a taurine concentration range of 1–40 $\mu$M, the platelets of the DI rats with chronic hypernatremia and hyperosmolality took up more taurine than those of the HZ and LE rats. After free water intake, the taurine uptake of the DI rats was approximately 60 % higher than that of the HZ and LE rats. Water deprivation induced a marked increase in taurine uptake of the DI rats, uptake being well over twofold that of the HZ and LE rats. Water deprivation produced a slight increase in taurine uptake of the LE rats, but the effect on the taurine uptake of the HZ rats was nearly negligible.

The uptake data were then analyzed and a Lineweaver-Burk plot was constructed to determine the kinetic parameters (Fig. 5B). The plots indicate that the $V_{max}$ of the DI

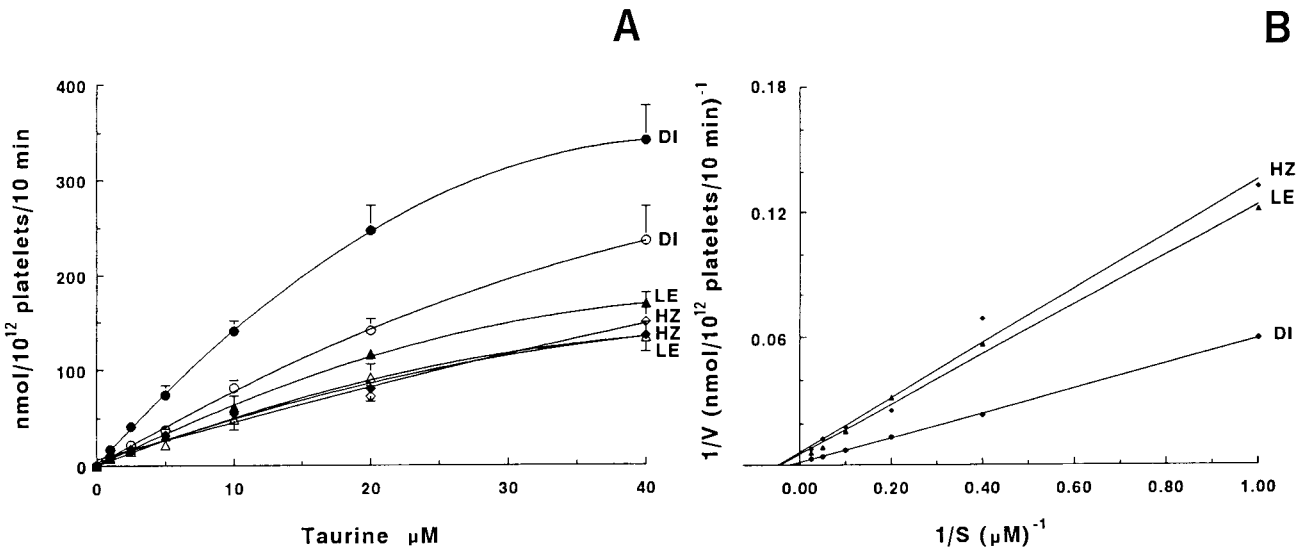


**Fig. 5** Taurine uptake by platelets of Brattleboro homozygotes (DI), Brattleboro heterozygotes (HZ) and Long Evans (LE) normal rats after free water intake for 24h (open symbols) and after water deprivation for 24h (filled symbols) at various concentrations of [$^3$H] taurine (**A**) and the Lineweaver-Burk reciprocal plots after water deprivation (**B**). The taurine uptake value at 37°C minus that at 0°C is shown. Each point represents the mean value ± S. E. (in 5B the mean value) of 5–10 experiments.

**Table 2** Kinetic constants of taurine uptake by platelets of Brattleboro homozygotes (DI), Brattleboro heterozygotes (HZ) and Long Evans (LE) normal rats after free water intake for 24h [water-control group (I)] and after water deprivation for 24h [water-deprived group (II)]

| | LE | HZ | DI |
|---|---|---|---|
| $V_{max}$ (nmol/$10^{12}$ platelets/10min) | | | |
| Free water intake (I) | 111 ± 11 | 127 ± 21 | 258 ± 32[b,c] |
| Water deprivation (II) | 182 ± 27 | 156 ± 28 | 479 ± 48[a,d] |
| $K_m$ ($\mu$M) | | | |
| Free water intake (I) | 17 ± 1 | 16 ± 4 | 25 ± 2[e] |
| Water deprivation (II) | 22 ± 3 | 23 ± 3 | 27 ± 2 |

Means ± S. E. from 5 to 10 animals are given.
Two-way ANOVA for $V_{max}$: group effect, $P < 0.001$; genotype effect, $P < 0.001$; and group × genotype interaction, $P < 0.05$.
Two-way ANOVA for $K_m$: genotype effect, $P < 0.05$.
Significance of differences: *a*) I *vs.* II, $P < 0.01$; *b*) DI *vs.* LE, $P < 0.001$; *c*) DI *vs.* HZ, $P < 0.05$; *d*) DI *vs.* HZ, LE $P < 0.001$; and *e*) DI *vs.* HZ, LE $P < 0.01$.

rats is about twofold the values of the LE and HZ rats after free water intake and threefold after water deprivation (Table 2). Water deprivation doubled the $V_{max}$ of the DI rats, without changing the $K_m$ value. Although the $K_m$ values of the LE and HZ rats are significantly lower after free water intake as compared to that of the DI rats, there is a trend toward an increased $K_m$ after water deprivation. Water deprivation increased the $V_{max}$ values of the LE and HZ rats approximately by 60 % and 20 %, respectively.

## Discussion

Valtin *et al.* reported in 1962 the discovery of a strain of rats with hereditary hypothalamic diabetes insipidus which was later named the Brattleboro strain (21). The

Brattleboro rats, homozygous for hereditary hypothalamic diabetes insipidus (DI), are totally deficient of vasopressin (22). Consequently, these animals exist in permanent state of water diuresis and must drink large quantities of water to maintain fluid balance. Despite enormous water drinking, the DI rats are mildly dehydrated most of the time. This is evidenced by a fairly elevated plasma sodium concentration · and osmolality compared to the Brattleboro heterozygotes (HZ) with 50 -60 % of normal vasopressin synthesis and normal rats of the Long Evans (LE) hooded strain. As expected, water deprivation for 24 h induced in the DI rats a drastic dehydration with exacerbated hypernatremia and hyperosmolality.

Plasma hypernatremia and hyperosmolality caused a substantial increase in platelet taurine of the DI rats. This adaptive increase in cell taurine in response to plasma hypernatremia and hyperosmolality may be due to stimulation of the taurine uptake system. Since the platelets contain large amounts of taurine (24) and they also accumulate taurine (15, 25), they may be a convenient model for studying taurine transport in anisosmotic conditions.

The taurine uptake by platelets of the Brattleboro rat exhibited a saturating tendency and was dependent on the incubation temperature, sodium concentration and osmolality of the plasma. Platelets from the DI rats with chronic hypernatremia and hyperosmolality showed consistently higher taurine uptake than that in platelets of the HZ and LE rats. Kinetic analysis indicated that the stimulation of the taurine uptake system of platelets by plasma hypernatremia and hyperosmolality occurs through a mechanism affecting $V_{max}$ rather than $K_m$. After free water intake, the $V_{max}$ of the taurine uptake in the DI rats was about two times higher than in the HZ and LE rats, and after water deprivation for 24 h it was about three times higher. In the DI rats, severe hypernatremia and hyperosmolality induced by water deprivation for 24 h increased the $V_{max}$ of the taurine uptake by twofold without changing $K_m$.

These results clearly indicate that the stimulation of the taurine uptake system is associated with and presumably accounts for the increased taurine content in platelets and probably also in other tissues of the DI rat with hypernatremia and hyperosmolality. Many cells and tissue preparations have been shown to accumulate taurine from the extracellular fluid by two processes: active uptake and passive diffusion. The active transport of taurine across the plasma membrane is a temperature- and sodium-dependent, carrier-mediated saturable process. This study shows that hypernatremia and hyperosmolality stimulate the taurine transport into platelets via an active component without any marked change in the diffusional component. The change observed in the active sodium-dependent component of taurine transport is probably not affected by modifications in the affinity of taurine for the platelet membrane carrier; this is indicated in the DI rats by the unchanged $K_m$ value after severe hypernatremia and hyperosmolality induced by water deprivation. In contrast, the high increase in $V_{max}$ suggests that an increase occurs in the number of functional carriers, or in the mobility of the taurine-carrier complex in the cell membrane. Very similar changes in the transport of taurine and also other organic osmolytes have been observed in many cells. Cultured astrocytes grown chronically in medium rendered hyperosmotic with sodium chloride showed an increase in the uptake of taurine (26). In the renal medullary cells grown in hypertonic media, an increased accumulation of taurine and other organic osmolytes, like glycerophosphorylcholine, betaine, inositol and sorbitol, has been demonstrated (27, 28). Moreover, increasing the concentration of sodium in the incubation medium enhanced taurine uptake in isolated cat retinas (29). Kinetic analysis indicates that these changes are due to increases in the $V_{max}$ of the transporters without any change in $K_m$.

An adaptive increase in platelet taurine concentration of the Brattleboro diabetes insipidus rat subsequent to plasma hypernatremia and hyperosmolality suggests that taurine plays a role in the maintenance of intracellular osmotic balance. The mechnism increasing the cell taurine content described in this work in platelets may operate also in intact tissues. It has long been known that high intracellular concentrations of electrolytes are toxic, inhibiting the function of proteins and enzymes (30, 31). By accumulating taurine and other organic osmolytes to balance the extracellular hyperosmolality, the cells are apparently protected from the perturbing effects of high intracellular concentrations of inorganic solutes.

Acknowledgments. We wish to thank Professor Heikki Vapaatalo for constructive criticism of the manuscript. This work was supported by the Research and Science Foundation of Farmos and the Sigrid Jusélius Foundation.

## References


1. Lasserre P and Gilles R: Modification of the amino acid pool in the parietal muscle of two euryhaline teleosts during osmotic adjustment. Experientia (1971) **27**, 1434-1435.
2. Gilles R: Intracellular organic osmotic effectors; in Mechanisms of Osmoregulation in Animals, Maintenance of Cell Volume, Gilles ed, John Wiley, New York (1979) pp111-154.
3. García JJ, Sánchez Olea R and Pasantes-Morales H: Taurine release associated to volume regulation in rabbit lymphocytes. J Cell Biochem (1991) **45**, 207 212.
4. Hoffmann EK and Hendil KB: The role of amino acids and taurine in isosmotic intracellular regulation in Ehrlich ascites mouse tumour cells. J Comp Physiol (1976) **108**, 279-286.
5. Lehmann A: Effects of microdialysis-perfusion with anisoosmotic media on extracellular amino acids in the rat hippocampus and skeletal muscle. J Neurochem (1989) **53**, 525-535.
6. Lehmann A, Carlström C, Nagelhus EA and Ottersen OP: Elevation of taurine in hippocampal extracellular fluid and cerebrospinal fluid of acutely hypoosmotic rats: Contribution by influx from blood? J Neurochem (1991) **56**, 690-697.
7. Pasantes-Morales H and Schousboe A: Volume regulation in astrocytes: A role for taurine as an osmoeffector. J Neurosci Res (1988) **20**, 505-509.
8. Pasantes-Morales H, García J and Sánchez-Olea R: Hyposmolarity-sensitive release of taurine and free amino acids from human lymphocytes. Biochem Pharmacol (1991) **41**, 303-307.
9. Solín JM, Herranz AS, Herreras O, Lerma J and Martín del Río R: Does taurine act as an osmoregulatory substance in the rat brain? Neurosci Lett (1988) **91**, 53-58.
10. Wade JV, Olson JP, Samson FE, Nelson SR and Pazdernik TL: A possible role for taurine in osmoregulation within the brain. J Neurochem (1988) **51**, 740-745.
11. Thurston JH, Hauhart RE and Dirgo JA: Taurine: A role in osmotic regulation of mammalian brain and possible clinical significance. Life Sci (1980) **26**, 1561-1568.
12. Thurston JH, Hauhart RE and Naccarato EF: Taurine: Possible role in osmotic regulation of mammalian heart. Science (1981) **214**, 1373 1374.
13. Nieminen M-L, Tuomisto L, Solatunturi E, Eriksson L and Paasonen MK: Taurine in the osmoregulation of the Brattleboro rat. Life Sci (1988) **42**, 2137 2143.
14. Paasonen MK, Penttilä O, Merikallio E, Siltanen P, Himberg JJ and Solatunturi E: Taurine in human auricular myocardium and blood platelets. Ann Clin Res (1982) **14**, 115-118.
15. Paasonen MK, Solatunturi E and Nieminen M-L: Blood platelets: Accumulation and release of taurine; in Taurine: Biological Actions and Clinical Perspectives, Oja, Ahtee, Kontro and Paasonen eds, Alan R.Liss, Inc., New York (1985) pp225-233.
16. Feissly R and Lüdin H: Microscopie par contrastes de phases. Rev Hémat (1949) **4**, 481-501.
17. Arora RC and Meltzer HY: A modified assay method for determining serotonin uptake in human platelets. Clin Chim Acta (1981) **112**, 225 -233.
18. Given MB and Longenecker GL: Characteristics of serotonin uptake and release by platelets; in The Platelets: Physiology and Pharmacology, Longenecker ed, Academic Press, Inc., London (1985) pp463-479.
19. Lineweaver H and Burk D: The determination of enzyme dissociation constants. J Am Chem Soc (1934) **56**, 658-666.
20. Tallarida RJ and Murray RB: Manual of Pharmacologic Calculations with Computer Programs, Springer-Verlag, New York Inc. (1987) pp61 -63.
21. Valtin H, Schroeder HA, Benirschke K and Sokol HW: Familial hypothalamic diabetes insipidus in rats. Nature (1962) **196**, 1109-1110.
22. Valtin H and Schroeder HA: Familial hypothalamic diabetes insipidus in rats (Brattleboro strain). Am J Physiol (1964) **206** (2), 425-430.
23. Valtin H: The discovery of the Brattleboro rat, recommended nomenclature, and the question of proper controls; in The Brattleboro Rat, Sokol and Valtin eds, The New York Academy of Sciences, New York (1982) pp1 9.
24. Soupart P: Free amino acids of blood and urine in the human, in Amino Acid Pools, Holden ed, Elsevier Publishing Co., New York (1962) pp220-262.
25. Ahtee L, Boullin DJ and Paasonen MK: Transport of taurine by normal human blood platelets. Br J Pharmacol (1974) **52**, 245-251.
26. Sánchez-Olea R, Morán J and Pasantes-Morales H: Changes in taurine transport evoked by hyperosmolarity in cultured astrocytes. J Neurosci Res (1992) **32**, 86-92.
27. Garcia-Perez A and Burg MB: Renal medullary organic osmolytes. Physiol Rev (1991) **71**, 1081-1115.
28. Uchida S, Nakanishi T, Kwon HM, Preston AS and Handler JS: Taurine behaves as an osmolyte in Madin-Darby canine kidney cells: Protection by polarized, regulated transport of taurine. J Clin Invest (1991) **88**, 656 662.
29. Schmidt SY: High-affinity uptake of [$^3$H] taurine in isolated cat retinas: Effects of $Na^+$ and $K^+$. Exp Eye Res (1980) **31**, 373 379.
30. Somero GN: Protons, osmolytes, and fitness of internal milieu for protein function. Am J Physiol (1986) **251**, R197-R213.
31. Yancey PH, Clark ME, Hand SC, Bowlus RD and Somero GN: Living with water stress: Evolution of osmolyte systems. Science (1982) **217**, 1214-1222.


Received May 15, 1995; accepted April 22, 1996.