Biochemical Pharmacology, Vol. 54, pp. 619–624, 1997.

ISSN 0006-2952/97/$17.00 + 0.00
PII S0006-2952(97)00213-X

Renal Excretory Responses to Saline Load in the Taurine-Depleted and the Taurine-Supplemented Rat

Mahmood S. Mozaffari, Junichi Azuma,† Champa Patel and Stephen W. Schaffer‡*

Department of Oral Biology/Pharmacology, Medical College of Georgia School of Dentistry, Augusta, GA 30912-1128, U.S.A.

ABSTRACT. Taurine is found in high concentrations in mammalian cells. Despite recognition of its role as an organic osmolyte in the kidney, information regarding its effects on renal fluid and electrolyte excretion is sparse. Therefore, the objective of the first series of experiments was to determine the effects of taurine depletion on renal excretory responses to a saline load. To induce taurine depletion, male Wistar-Kyoto (WKY) rats were treated with tap water containing 3% β-alanine for 3 weeks. Taurine depletion reduced the initial rates of fluid and sodium excretion after an intravenous saline load. This effect was attributed to taurine depletion since maintenance of the taurine-depleted rats on tap water for 2 days to remove the effects of β-alanine yielded the same pattern as the taurine-depleted rats exposed to β-alanine at the time of the experiment. Nonetheless, rats exposed to short-term β-alanine treatment, which has no influence on kidney taurine content, demonstrated a larger (~25%) natriuretic but not diuretic response to the isotonic saline load than either the control or taurine-depleted rats. These data suggest that β-alanine-induced inhibition of tubular reabsorption of taurine may result in subsequent excretion of taurine with attendant natriuresis early in the course of β-alanine treatment. We also tested the hypothesis that taurine potentiates the renal excretory responses to an isotonic saline load in WKY rats. Inclusion of taurine in the infusate significantly increased natriuresis and diuresis after a saline load. This effect was greater in animals fed a basal than a high NaCl diet. Our data support a role for taurine as a natriuretic and diuretic agent. BIOCHEM PHARMACOL 54;5:619–624, 1997. © 1997 Elsevier Science Inc.

KEY WORDS. Renal; taurine; β-alanine; natriuretic; diuretic

Taurine (2-aminoethanesulfonic acid) is the most abundant free amino acid in the kidney [1]. Numerous physiological roles have been attributed to taurine [1–5] including an osmoregulatory function in the kidney [1]. However, relatively little is known about its effect on fluid and sodium homeostasis.

A major renal mechanism contributing to the regulation of sodium and fluid excretion is the counter-current multiplier system. This system establishes an ionic gradient along the corticomedullary axis, which results in a high concentration of interstitial sodium. This high interstitial sodium concentration (i.e. osmolality) not only promotes reabsorption of fluid from the renal tubular system, thereby concentrating urine, but it also exposes the renal tubular cells to a high osmotic stress. The renal tubular cells, in turn, adjust to this environment by accumulating the organic osmolyte taurine [6–8]. Therefore, a reduction in intracellular taurine concentration would be expected to adversely affect the ability of the kidney to regulate sodium and fluid excretion. To test this idea, the present study examined whether depletion of endogenous taurine stores in rats affects renal excretory responses induced by the administration of a saline load.

MATERIALS AND METHODS

Taurine Depletion Protocol

Six-week-old male WKY§ rats were obtained from Harlan Laboratories (Indianapolis, IN). All rats were maintained two per cage at constant humidity (60 ± 5%), temperature (24 ± 1°), and light cycle (6:00 a.m. to 6:00 p.m.). Two days after arrival, the animals were randomly assigned to two groups: one group received drinking water containing 3% β-alanine (taurine-depleted group; N = 4) and the control group (N = 4) received only tap water (Fig. 1). β-Alanine inhibits cellular uptake of taurine, and its administration in the drinking water is an acceptable approach to reduce endogenous taurine stores [9–11]. Food and drinking fluid were available *ad lib.* throughout the study.

Two days prior to examining the natriuretic and diuretic responses to a saline load, each rat was instrumented under

* Corresponding author: Tel. (706) 721-3181; FAX (706) 721-6276.
† Current address: Department of Clinical Evaluation of Medicines and Therapeutics, Osaka University, Osaka, Japan.
‡ Current address: Department of Pharmacology, College of Medicine, University of South Alabama, Mobile, AL 36688.
§ *Abbreviations:* WKY, Wistar Kyoto; and ANP, atrial natriuretic peptide.
Received 13 January 1997; accepted 19 March 1997.

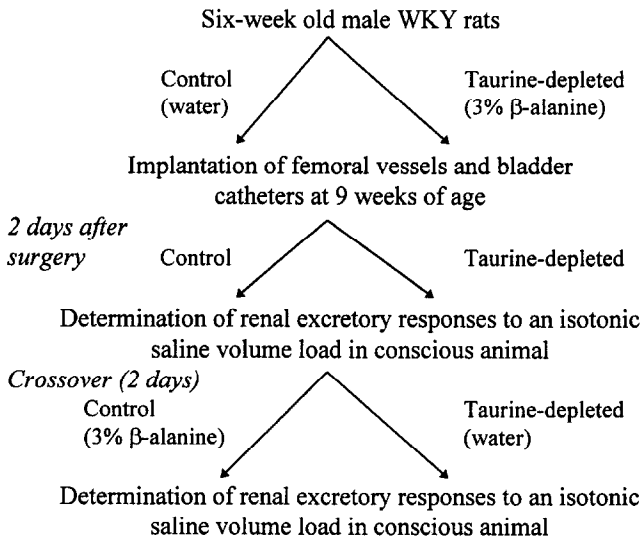

FIG. 1. General experimental protocol for control and taurine-depleted rats. All rats were maintained on tap water containing either no addition (control) or 3% β-alanine (taurine-depleted) for 3 weeks. After completing the surgery, the first saline infusion protocol was performed. Thereafter, a crossover was initiated during which the control animals received 3% β-alanine while the taurine-depleted rats were given tap water for 2 days. The experiment concluded with a re-determination of renal excretory responses to a saline load.

ether anesthesia with femoral arterial and venous catheters (PE-10 fused with PE-50), and a bladder catheter as described previously [12]. On the day of the experiment, each rat was placed in an environmental conditioning unit (Braintree, Braintree, MA); all rats had been conditioned to the units for 5 days prior to testing. After flushing the arterial and venous catheters with 0.3 mL of isotonic saline containing 5 U/mL of heparin, the recording of mean arterial pressure and heart rate was initiated. A baseline urine sample (30 min) was obtained prior to intravenous infusion of isotonic saline (equivalent to 5% body weight at the rate of 0.5 mL/min). After initiation of the isotonic saline infusion, urine samples were collected at 15, 30, 60, and 90 min, and animals were returned to their home cages. Urine volume was determined, and urinary Na^+ concentration was measured by flame photometry.

After 3 weeks on β-alanine or tap water, the same rats were subjected to a crossover protocol in which the taurine-depleted group was maintained for 2 days on normal tap water while the control group was treated with 3% β-alanine for 2 days. They were then subjected to an i.v. isotonic saline load as described above (see Fig. 1 for a flow chart describing the protocol). Urine samples were collected and both volume and Na^+ concentration determined.

The extent of β-alanine-induced taurine depletion was determined in a subset of animals that were not used in the infusion studies. This was necessary in order to determine the magnitude of taurine-depletion without the confounding influence of the crossover protocol. This is an important consideration since we have demonstrated that taurine repletion of β-alanine-treated rats restores renal taurine content to the level of untreated control rats (data not published). Kidneys from control (N = 4) and taurine-depleted WKY rats (N = 4) that were on 3% β-alanine for 3 weeks were removed from the animals, rinsed in isotonic saline, weighed, and stored in liquid nitrogen at −70° until assayed. Renal taurine content was determined using extracts of freeze-dried tissue as we have described previously [10].

Dietary NaCl Treatment

Ten-week-old male WKY rats were obtained from Harlan Laboratories. Two days after arrival, the animals either remained on a basal NaCl diet (1%; N = 9) or were placed on a high NaCl diet (8%; N = 7) and housed as described above. Dietary NaCl excess did not affect significantly body weight, mean arterial pressure, or heart rate in WKY rats [12]. Eight days after arrival, each rat was implanted with femoral vessels and bladder catheters. Two days after surgery, the arterial and venous lines were flushed with 0.3 mL of isotonic saline containing 5 U/mL of heparin, and the mean arterial pressure and heart rate of the rats were recorded. A basal urine sample was collected for 30 min prior to the i.v. infusion of an isotonic saline load equivalent to 2.5% body weight of the animal at the rate of 0.5 mL/min (phase I). Urine was collected for 45 min at 15-min intervals. Thereafter, each rat was reinfused with isotonic saline containing 1% taurine (2.5% body weight; 0.5 mL/min; phase II), and urine samples were collected for three consecutive 15-min intervals. At the conclusion of the experiment, the animals were returned to their home cages. Two days later, each rat was given two successive saline loads according to the above protocol; the second saline load did not contain taurine. The data served as a control by evaluating the effects of two consecutive saline loads on natriuresis and diuresis. At the conclusion of the infusion studies, the animals were killed with urethane.

Statistics Analysis

All data were analyzed by the analysis of variance (significance criteria of $P < 0.05$) with appropriate post-hoc tests (Newman–Keuls) to determine main effects and interactions.

RESULTS

To produce a taurine-depleted animal, rats were maintained on tap water containing 3% β-alanine for 3 weeks. Although this treatment had no effect on body weight, mean arterial pressure, or heart rate (Table 1), it resulted in a significant reduction in renal tissue taurine content (44.5 ± 1.3 vs 25.3 ± 3.4 μmol/g dry weight for control and 3% β-alanine-treated rats, respectively; $P < 0.05$). Three weeks of β-alanine treatment also increased the

TABLE 1. Body weight, mean arterial pressure, and heart rate of control and taurine-depleted rats

	Body weight (g)	Mean arterial pressure (mm Hg)	Heart rate (beats/min)
Control	211 ± 6	126 ± 4	425 ± 20
Taurine-depleted	207 ± 3	129 ± 4	415 ± 19

Values are means ± SEM, N = 4 per group.

wet/dry weight ratio of renal tissue (4.13 ± 0.15 vs 3.53 ± 0.11).

Figure 2 reveals that baseline urinary output and Na^+ excretion rates were similar between the untreated control (maintained on tap water) and the taurine-depleted group (maintained on β-alanine). Infusion of an isotonic saline load resulted in a rapid, significant diuresis and natriuresis in both groups. In these rats, the peak in urinary flow rate and Na^+ excretion rate occurred 30 min after initiation of saline infusion. Thereafter, urinary fluid and Na^+ excretion returned towards the baseline level. Interestingly, renal excretion of fluid and sodium was higher in the control group than in the taurine-depleted group 15 min after initiation of the saline load (Fig. 2).

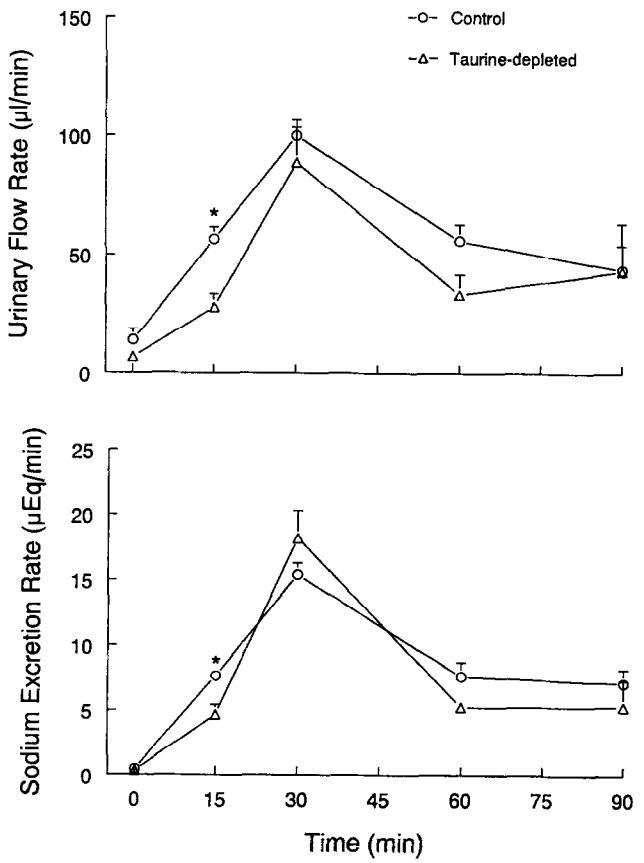

FIG. 2. Urinary flow rate (top panel) and sodium excretion rate (bottom panel) in taurine-depleted and control WKY rats. Values are the means ± SEM of data from four control and four taurine-depleted rats. Key: (*) $P < 0.05$ compared with other groups.

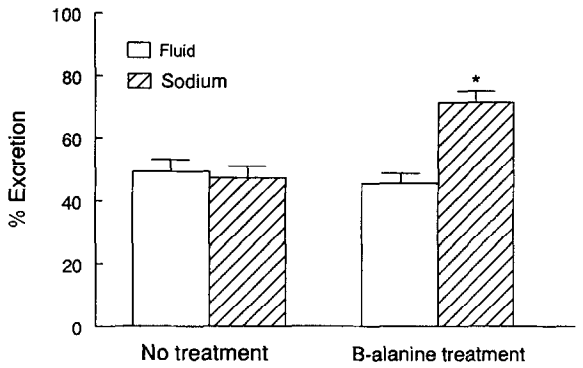

FIG. 3. Effect of short-term (2 days) 3% β-alanine treatment on the percent of the saline load excreted 90 min after initiation of a saline load. Values are the means ± SEM of four rats treated with β-alanine and four rats treated with tap water. Key: (*) $P < 0.05$ compared with no treatment.

Since β-alanine is osmotically active and mediates certain biological effects [11, 13, 14], a crossover study was performed to evaluate the effect of β-alanine independent of cellular taurine content. In this study, control untreated rats were maintained for 2 days on water containing 3% β-alanine (short-term β-alanine exposure) while rats treated for 3 weeks with 3% β-alanine were placed on tap water for 2 days. The 2-day treatment with β-alanine caused no significant change in renal taurine content while rats maintained for 3 weeks on 3% β-alanine followed by 2 days on normal tap water exhibited reduced renal taurine levels. These control animals were then subjected to the standard saline loading protocol. Interestingly, the diuretic and natriuretic responses of the taurine-depleted group were not altered significantly by changing the drinking fluid from 3% β-alanine to normal tap water for 2 days (data not shown). Similarly, short-term β-alanine exposure had no influence on fluid excretion. However, the sodium excretion rate was significantly ($P < 0.05$) higher after, compared with before, short-term β-alanine exposure (Fig. 3). Thus, while taurine depletion (long-term β-alanine treatment) significantly reduced fluid excretion (Fig. 2), short-term β-alanine treatment increased sodium excretion without altering fluid excretion (Fig. 3). Potassium excretion was similar between all groups, either before or after the crossover protocol (data not shown).

Since differential effects were noted between β-alanine-treated animals containing lower intracellular taurine levels and normal taurine content, the effect of acute taurine exposure was examined. Figure 4 reveals an effect of acute taurine exposure on both fluid and sodium excretion following a saline load. In this experiment, the animals were subjected to two consecutive saline challenges. The initial saline challenge (phase I) led to a smaller saline-induced stimulation in fluid and sodium excretion than the second challenge (phase II; Fig. 4). Inclusion of taurine in the infusate potentiated the diuretic and natriuretic responses to the saline load during phase II, compared with phase I (saline only, Fig. 4), further increasing the differences between the phase I and phase II responses.

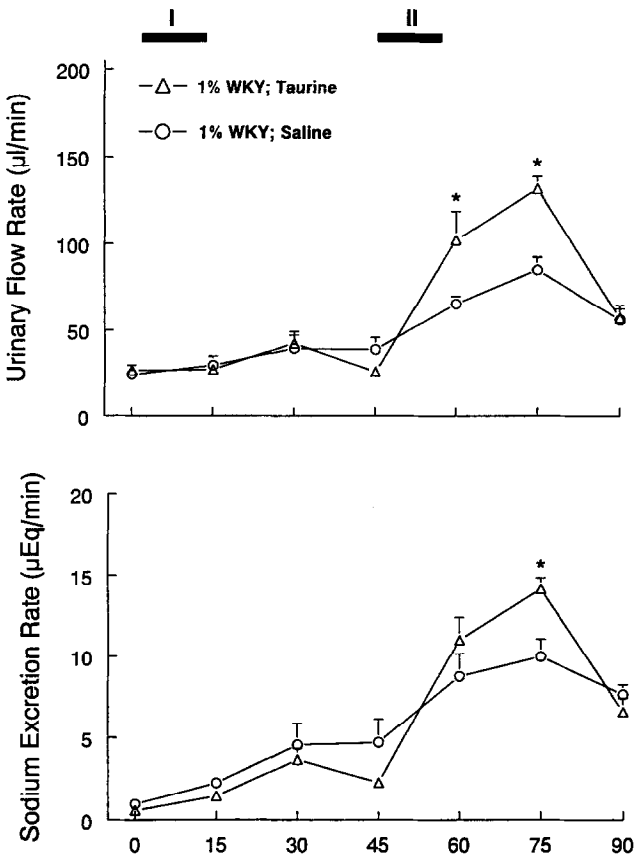

FIG. 4. Urinary flow rate (top panel) and sodium excretion rate (bottom panel) in response to an initial (phase I) and a second (phase II) saline infusion protocol. Values are the means ± SEM of nine rats. Key: (*) $P < 0.05$ compared with the saline group at the same time point.

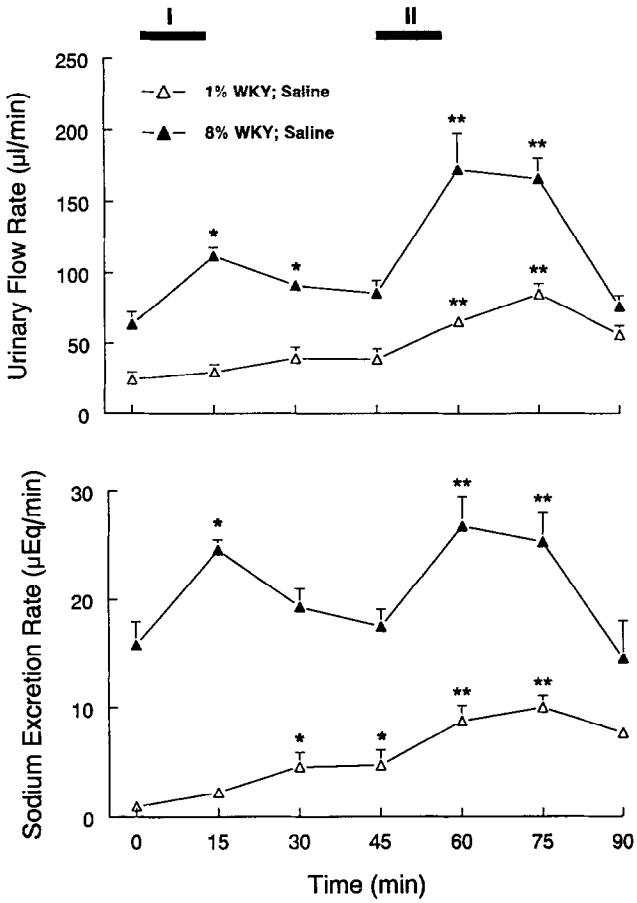

FIG. 5. Effect of dietary NaCl excess on urinary flow rate (top panel) and sodium excretion rate (bottom panel) in response to two successive saline loads (I and II). Values are the means ± SEM of renal function in nine 1% and seven 8% NaCl diet rats. Key: (*) $P < 0.05$ compared with the zero minute time point in the same group; and (**) $P < 0.05$ compared with the 45-min time point in the same group.

Another procedure commonly used to alter renal fluid and sodium excretion rates is to elevate dietary NaCl content [12]. This effect is seen in Fig. 5 for rats that were fed 1% (basal) or 8% (high) NaCl diet for 8 days prior to determining renal responses to an acute intravenous saline load. Baseline fluid and sodium excretion rates were significantly ($P < 0.05$) higher in the animals maintained on the high, compared with the basal NaCl diet. Infusion of the initial isotonic saline load (phase I) resulted in a greater increase in both diuresis and natriuresis in rats fed the high, compared with the basal, NaCl diet. Following cessation of the saline infusion, the diuretic and natriuretic responses slowly returned towards the baseline level in the high NaCl diet group, but there was a slight shift towards higher excretion levels in the basal NaCl diet group (Fig. 5). The second saline challenge (phase II) resulted in significant increases in fluid and Na^+ excretion, which were greater in animals maintained on the high, compared with the basal, NaCl diet.

Despite the differences in sodium and fluid excretion between the animals fed the 8% and the 1% NaCl diets, the response to acute taurine exposure was qualitatively similar. Figure 6 summarizes the natriuretic and diuretic responses to a saline load expressed as the percent change in the fluid and Na^+ excretion in phase II (46–90 min) compared with phase I (0–45 min). Acute taurine exposure elevated both fluid and sodium excretion in rats fed either the high or the basal NaCl diet, although the elevation in fluid excretion by acute taurine exposure was not significant in the high NaCl group. The differences in the renal responses to the high NaCl diet could not be attributed to either changes in mean arterial pressure or heart rate since these parameters were not affected by dietary NaCl excess. Moreover, the infusion protocol also had no significant effect on either hemodynamic parameter (data not shown).

DISCUSSION

The present study indicates that chronic β-alanine treatment resulting in significant taurine depletion slightly reduces urinary output in response to a saline load in WKY rats. By contrast, short-term exposure (i.e. 2 days of treatment) of rats to β-alanine significantly increases the natriuretic response to a saline load. Since renal taurine transport occurs via a sodium-gradient-dependent mechanism

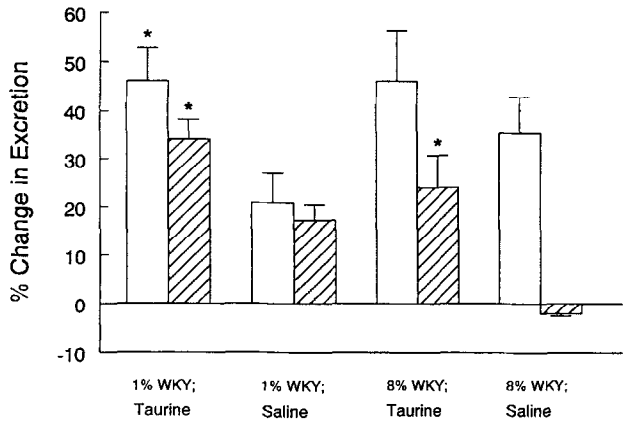

FIG. 6. Effect of taurine and dietary NaCl excess on phase I and phase II renal excretion of fluid and sodium. Data are expressed as the percent difference between the renal response to an initial (phase I; 0–45 min) and a second (phase II; 46–90 min) saline load. Open and hatched bars represent fluid and sodium excretion, respectively. Values are the means ± SEM of renal function in nine 1% and seven 8% NaCl diet rats. Key: (*) $P < 0.05$ compared with the saline-treated group on the same diet.

[8] and β-alanine inhibits renal reabsorption of taurine, the natriuretic response to short-term β-alanine treatment could be secondary to β-alanine-induced taurinuria. This possibility was tested directly by determination of renal responses to a saline load in the presence and absence of taurine in the infusate. As shown in Fig. 6, acute administration of taurine significantly increased the diuretic and natriuretic responses to a saline load, and these effects were more prominent in animals maintained on a basal, compared with a high NaCl diet. Our data are in general agreement with the hypothesis that taurine affects renal fluid and electrolyte homeostasis and corroborates the findings of Gentile and colleagues [15] who reported that taurine causes significant improvement in renal excretory responses in cirrhotic patients with ascites.

Taurine is believed to function as an osmoregulator in the kidney [1]. This concept originates from the recognition that renal tubular cells are exposed to an increased external salt gradient along the corticomedullary axis. Therefore, an intracellular buildup of taurine in the tubular cells serves to maintain intracellular osmolality. β-Alanine treatment can be expected to elicit a physiological adaptive mechanism(s) related to the reduction in renal taurine content. One possibility could involve a greater rate of synthesis and/or sequestration of other organic osmolytes, i.e. glycine and betaine in lieu of taurine. Alternatively, there could be an adjustment of the interstitial osmolality, which would involve greater tubular reabsorption of fluid into the interstitium, thereby reducing interstitial osmolality. An investigation of the renal mechanism(s) of physiological adaptation to taurine depletion was not the subject of the current study. However, it is noteworthy that the wet/dry weight ratio of the kidneys from taurine-depleted rats was significantly higher than that of the control rats, indicating greater renal tissue water content. If β-alanine-induced taurine depletion results in greater tubular reabsorption of water (and attendant sodium reabsorption), this could explain, at least in part, the reduction in the diuretic response of taurine-depleted rats to a saline load. Since β-alanine-induced taurine depletion did not affect systemic arterial pressure, it is unlikely that a pressure-diuresis-natriuresis mechanism contributed significantly to the renal responses of the β-alanine-treated rats.

Another noteworthy finding of the present study is the demonstration that when WKY rats were given two consecutive saline loads, the natriuretic and diuretic responses to the second stimulus were stronger than the previous one (see Figs. 4 and 5). Despite the larger diuresis and natriuresis during the second saline load, taurine-induced potentiation of these responses still persisted during the second phase of the infusion protocol. Isotonic saline-induced renal excretions of fluid and sodium are mediated by integrated responses that involve decreased renal sympathetic nerve activity, suppression of the renin-angiotensin-aldosterone axis [see Ref. 16], and increased secretion of ANP [17–19]. Therefore, a likely contributor to the augmented renal responses to the second saline load, as well as the taurine-induced modulation of these responses, is ANP. In support of this view, Dlouha and McBroom [20] demonstrated that atrial extracts from taurine-treated hamsters increase renal excretion of Na^+ and fluid in rat, suggesting an association between taurine and ANP.

Taurine-induced potentiation of diuresis and natriuresis is more prominent in animals fed a basal, compared with a high, NaCl diet. Dietary NaCl supplementation in WKY rats is accompanied by a significant increase in basal excretion of Na^+ and fluid, as well as greater diuretic and natriuretic responses to a saline load [12]. Therefore, it is likely that the taurine-mediated improvement in dispensing of the saline load is partially masked in the animals maintained on a high NaCl diet. This accounts for the smaller taurine effect in the high NaCl diet group (Fig. 6).

The demonstration of the influence of taurine on renal handling of fluid and Na^+ should be considered in the overall context of the effects of taurine on the cardiovascular system. In a recent double-blind randomized crossover clinical trial, Azuma [21] found that taurine therapy benefited patients with congestive heart failure. The same group [22] also found that taurine treatment decreases the cumulative mortality rate in rabbits with congestive heart failure induced by aortic regurgitation. Two mechanisms may contribute to these phenomena. First, taurine has been shown to mediate a positive inotropic effect in rabbits with experimentally induced chronic aortic regurgitation [23]. This action of taurine represents a direct effect on the compromised heart, either involving improved calcium transport [24] or increased sensitivity of the muscle proteins to calcium [25]. The present study raises another potential mechanism of taurine action. By promoting diuresis and natriuresis, taurine therapy may decrease sodium and fluid retention and minimize the expansion of extracellular fluid and edema formation. Thus, taurine may combine the

beneficial effects of diuretics and positive inotropic agents, thereby serving as an effective drug in the treatment of congestive heart failure.

In conclusion, taurine depletion mediated modest changes in renal responses to a saline load. However, acute administration of taurine caused significant increases in the diuretic and natriuretic responses to an isotonic saline volume load. Taurine is a safe compound with no adverse metabolic effects. Therefore, further investigation of its potential usefulness in conditions with compromised cardiovascular and renal function is warranted.

This study was supported, in part, by a grant from the American Heart Association (Georgia affiliate) and the Taisho Pharmaceutical Co. of Japan.

References

1. Nakanishi T, Takamitsu Y and Sugita M, Role of taurine in the kidney: Osmoregulatory taurine accumulation in renal medulla. In: *Taurine in Health and Disease* (Eds. Huxtable RJ and Michalk D), pp. 139–148. Plenum Press, New York, 1994.
2. Huxtable RJ, Physiological actions of taurine. *Physiol Rev* **72:** 101–163, 1992.
3. Schaffer SW, Azuma J and Madura JD, Mechanisms underlying taurine-mediated alterations in membrane function. *Amino Acids* **8:** 231–246, 1995.
4. Sturman JA, Taurine in development. *Physiol Rev* **73:** 119–147, 1993.
5. Chesney RW, Taurine: Its biological role and clinical implications. *Adv Pediatr* **32:** 1–42, 1985.
6. Amiry-Moghaddam M, Nagelhus E and Ottersen OP, Light- and electronmicroscopic distribution of taurine, an organic osmolyte, in rat renal tubule cells. *Kidney Int* **45:** 10–22, 1994.
7. Trachtman H, Lu P and Sturman JA, Immunohistochemical localization of taurine in rat renal tissue: Studies in experimental disease states. *J Histochem Cytochem* **41:** 1209–1216, 1993.
8. Zelikovic I, Stejskal-Lorenz E, Lohstroh P, Budreau A and Chesney RW, Anion dependence of taurine transport by rat renal brush border membrane vesicles. *Am J Physiol* **256:** F646–F655, 1989.
9. Jones DP, Miller LA and Chesney RW, Adaptive regulation of taurine transport in two continuous renal epithelial cell lines. *Kidney Int* **38:** 219–226, 1990.
10. Mozaffari MS, Tan BH, Lucia MA and Schaffer SW, Effect of drug-induced taurine depletion on cardiac contractility and metabolism. *Biochem Pharmacol* **35:** 985–989, 1986.
11. Lu P, Xu W and Sturman JA, Dietary β-alanine results in taurine depletion and cerebellar damage in adult cats. *J Neurosci Res* **43:** 112–119, 1996.
12. Mozaffari MS, Jirakulsomchok S, Shao ZH and Wyss JM, High-NaCl diets increase natriuretic and diuretic responses in salt-resistant but not salt-sensitive SHR. *Am J Physiol* **260:** F890–F897, 1991.
13. Lake N and DeMarte L, Effects of β-alanine treatment on taurine and DNA content of the rat heart and retina. *Neurochem Res* **13:** 1003–1006, 1988.
14. Chovan JP, Kulakowski EC, Sheakowski S and Schaffer SW, Calcium regulation by the low affinity taurine binding site of cardiac sarcolemma. *Mol Pharmacol* **17:** 295–300, 1980.
15. Gentile S, Bologna E, Terracina D and Angelico M, Taurine-induced diuresis and natriuresis in cirrhotic patients with ascites. *Life Sci* **54:** 1585–1593, 1994.
16. Hass JA, Granger JP and Knox FG, Effect of intrarenal volume expansion on proximal sodium reabsorption. *Am J Physiol* **255:** F1178–F1182, 1988.
17. Watenpaugh DE, Yancy CW, Buckey JC, Lane LD, Hargens AR and Blomqvist G, Role of atrial natriuretic peptide in systemic responses to acute isotonic volume expansion. *J Appl Physiol* **73:** 1218–1226, 1992.
18. Shinohara M, Nonoguchi H, Ujiie K, Terada Y, Owada A, Tomita K and Marumo F, Effects of hyperosmolality on ANP-stimulated cGMP generation in rat inner medullary collecting duct. *Kidney Int* **45:** 1362–1368, 1994.
19. Knepper MA, Lankford SP and Terada Y, Renal tubular actions of ANF. *Can J Physiol Pharmacol* **69:** 1537–1545, 1991.
20. Dlouha H and McBroom MJ, Atrial natriuretic factor in taurine-treated normal and cardiomyopathic hamsters. *Proc Soc Exp Biol Med* **181:** 411–415, 1986.
21. Azuma J, Long term effect of taurine in congestive heart failure: Preliminary report. In: *Taurine in Health and Disease* (Eds. Huxtable RJ and Michalk D), pp. 425–433. Plenum Press, New York, 1994.
22. Takihara K, Azuma J, Awata N, Ohta H, Hamaguchi T, Sawamura A, Tanaka Y, Kishimoto S and Sperelakis N, Beneficial effect of taurine in rabbits with chronic congestive heart failure. *Am Heart J* **112:** 1278–1284, 1986.
23. Awata N, Azuma J, Hamaguchi T, Tanaka Y, Ohta H, Takihara K, Harada H, Sawamura A and Kishimoto S, Acute hemodynamic effect of taurine on hearts *in vivo* with normal and depressed myocardial function. *Cardiovasc Res* **21:** 241–247, 1987.
24. Schaffer SW, Ballard C and Azuma J, Mechanisms underlying physiological and pharmacological actions of taurine on myocardial calcium transport. In: *Taurine in Health and Disease* (Eds. Huxtable RJ and Michalk D), pp. 171–180. Plenum Press, New York, 1994.
25. Steele DS, Smith GL and Miller DJ, The effects of taurine on Ca^{2+} uptake by the sarcoplasmic reticulum and Ca^{2+} sensitivity of chemically skinned rat heart. *J Physiol (Lond)* **422:** 499–511, 1990.