Pharmacology Biochemistry and Behavior, Vol. 44, pp. 263-266, 1993
Printed in the U.S.A.

0091-3057/93 $6.00 + .00

Taurine and Ethanol-Induced Conditioned Taste Aversion

C. M. G. ARAGON AND Z. AMIT[1]

Center for Studies in Behavioral Neurobiology, Concordia University, 1455 de Maisonneuve Boulevard West, Montreal, Quebec H3G 1M8, Canada

Received 28 April 1992

ARAGON, C. M. G. AND Z. AMIT. *Taurine and ethanol-induced conditioned taste aversion.* PHARMACOL BIOCHEM BEHAV **44**(2) 263-266, 1993. – It has been reported that acute, simultaneous injections of taurine and ethanol were effective in reducing ethanol-induced locomotor activity and sleep time. The possible involvement of taurine administration in ethanol-induced conditioned taste aversion (CTA) was investigated. The results obtained in the present study following simultaneous administration of taurine (45 mg/kg) and ethanol (0.8, 1.2, and 1.6 g/kg) demonstrate a significant interaction between taurine and ethanol in their effect on ethanol-induced CTA in rats. This interaction was biphasic in nature and dependent upon the specific dose of ethanol. At the lowest ethanol dose (0.8 g/kg), which in itself resulted in a marginal CTA, taurine significantly enhanced the CTA induced by this dose. The intermediate ethanol dose of 1.2 g/kg produced a significant CTA. This CTA was blocked by administration of taurine. Finally, the CTA produced by the high dose of ethanol (1.6 g/kg) was not affected by administration of taurine. Taurine by itself does not produce a CTA. Peripheral levels of ethanol were ethanol dose dependent and the same in all animals regardless of treatment, indicating taurine had no effects on plasma ethanol levels. These data are similar to those obtained by earlier studies on the effects of taurine on ethanol-induced motor activity in mice. The present results support the findings reported by other investigators that taurine administration exerts a significant effect on ethanol-induced behaviors.

Ethanol Taurine Conditioned taste aversion

ONE of the central research topics during the past two decades within the broad field of alcohol studies has been the attempt to identify the central mechanisms underlying alcohol's effects on behavior. Indeed, these efforts have clearly been predicted on the assumption that the data that will accumulate as a result of these studies will contribute to our understanding of the development of alcoholism and in particular the propensity of organisms to voluntarily ingest alcohol.

A more specific group of studies within this larger body of evidence attempted to examine the possible role of the amino acid taurine in the control of one or more of ethanol's effects on behavior. The initial study in this area investigated the effects of taurine on ethanol-induced narcosis (4,10,13). These investigators reported that sleep time resulting from administration of large doses of ethanol was reduced following injections of taurine. The decrease in sleep time was reported to be substantially greater with taurine compared to nine other amino acids administered in similar concentrations and in an identical paradigm to that used with taurine (10). Further, it appeared, at least initially, that this effect of taurine was behavior specific because taurine did not antagonize the effects of ethanol on body temperature or susceptibility to seizures (4).

These initial observations that taurine may have a specific effect on ethanol-related behaviors raised some obvious questions concerning the possible mechanism underlying the observed effects. Several such putative mechanisms have actually been proposed. Naturally, given the weight of the evidence most of the suggestions concerning a taurine-related mechanism focused on its ability to antagonize ethanol-induced narcosis. In this context, it was proposed that taurine may be acting as a general neuromodulator. This notion is in line with reports suggesting that taurine may be functioning within the CNS as an inhibitory neurotransmitter (7,8,12,18). Taurine has also been proposed, on the basis of data reported by Huxable and Bressler (9), as a membrane stabilizer. These researchers found, using a sarcoplasmic reticulum preparation from a rat skeletal muscle, that taurine caused an increase in the rate of calcium oxalate uptake, as well as an increase in the total calcium sequestered and a decrease in the rate of calcium transport after treatment of the preparation with phospholipase C. A third possible mechanism putatively underlying the effects of taurine on ethanol-induced sleep has been proposed by Iida and Hikichi (10), who suggested that ethanol or acetaldehyde may interact with taurine to form a compound capable of exerting direct behavioral effects or increasing the metabolic rate of ethanol.

Finally, a role for taurine as a possible activator of the

[1] To whom requests for reprints should be addressed.

enzyme aldehyde dehydrogenase has also been proposed as underlying the observed ethanol–taurine interaction (15,16). In line with this proposal, it was found that following administration of taurine a significant reduction in blood acetaldehyde levels in rats and "flushing" in humans was observed (15). On the basis of these observations, it was suggested that taurine, in the above-mentioned study, may have accelerated the oxidation of acetaldehyde by activating aldehyde dehydrogenase.

So far, most of the data relating taurine to ethanol's effects came from experiments studying movement (3) or lack of it during sleep (4,10,13). However, given the earlier finding (4) that taurine did not affect ethanol-induced changes in body temperature or seizure susceptibility one had to consider the possibility that the taurine–ethanol interaction is underlying a narrow band of ethanol-related behavioral variables. The present study is an attempt to examine this possibility by extending the range of behavioral variables studied. More specifically, we designed this study to examine the possible effects of taurine on ethanol-induced conditioned taste aversion (CTA).

The ability of ethanol in subanesthetic doses to induce a CTA is well established (1,2,6) and is a sensitive, dose-dependent paradigm to study ethanol effects on behavior. Because a CTA was demonstrated in a sleeping animal preparation (5), it is clear that this paradigm is, at most, only minimally dependent upon central mechanisms implicated in the regulation of motor movement. Given these properties of the CTA paradigm, we felt that its usage in the present experiment may contribute valuable data on the extent of taurine's involvement in mediating ethanol-induced changes in behavior.

METHOD

Subjects

Seventy-four male Long-Evans rats (Charles River, Canada) were used in this study. They weighed 250–310 g at the beginning of the experiment. Animals were housed individually in stainless steel cages with free access to food and water prior to the beginning of testing. The room in which animals were housed had air and humidity control and was illuminated on a 12 L : 12 D cycle.

Drugs

Taurine, purchased from Sigma Chemical Co. (St. Louis, MO), was dissolved in distilled water in a concentration of 20 mg/10 ml. Ethanol solutions in a concentration of 20% v/v were prepared by mixing 95% ethanol in tapwater.

Procedure

Following 1-week adaptation to the laboratory conditions, animals were placed on a 23-h 40-min water deprivation schedule. Water was presented to animals in stoppered plastic test tubes fitted with stainless steel spouts containing double ball bearings. The spouts were inserted through the wire mesh in front of the cages. Water consumption was measured to the nearest milliliter. The eighth day following the onset of the deprivation schedule constituted "pairing day 1." On this day, rats were presented with a 0.1% v/v sodium saccharin solution instead of the water during the 20 min fluid was presented to them. This saccharin solution represented the novel tasting fluid and the conditioned stimulus (CS), whose consumption in the CTA paradigm constituted the dependent variable. Following the 20-min drinking session on pairing day 1, rats were randomly assigned to one of eight groups. The treatment given to the various groups is described below, however; all groups received, immediately following the drinking session, two IP injections in a drug dose that varied according to the group.

Group S-E-0.8 (n = 8) received an injection of saline followed by an injection of ethanol in a dose of 0.8 g/kg.
Group S-E-1.2 (n = 8) received an injection of saline followed by an injection of ethanol in a dose of 1.2 g/kg.
Group S-E-1.6 (n = 8) received an injection of saline followed by an injection of ethanol in a dose of 1.6 g/kg.
Group T-E-0.8 (n = 8) received an injection of taurine in a dose of 45 mg/kg followed by an injection of ethanol in a dose of 0.8 g/kg.
Group T-E-1.2 (n = 8) received an injection of taurine in a dose of 45 mg/kg followed by an injection of ethanol in a dose of 1.2 g/kg.
Group T-E-1.6 (n = 8) received an injection of taurine in a dose of 45 mg/kg followed by an injection of ethanol in a dose of 1.6 g/kg.
Group S-S (n = 13), one of two control groups used in this study, received two injections of saline immediately following the end of the drinking session.
Group T-S (n = 13), the second control group, received an injection of taurine (45 mg/kg) followed by an injection of saline.

During a 5-day period following pairing day 1, the rats were, once again, presented with tapwater during the 20-min daily drinking sessions. Day 14 from the start of the fluid deprivation schedule constituted pairing day 2. On this day, rats in their various groups were treated in a manner identical to that employed on pairing day 1. Following pairing day 2, once again rats were presented with tapwater during their 20-min daily drinking session for a period of 5 days. Finally, day 20 from the start of the experiment constituted "test day 1." On this day, rats were presented with a saccharin solution during their 20-min daily drinking session but no injections of any kind were administered following the drinking. A final 5-day period during which tapwater was presented to animals followed test day 1. Day 26 from the start of the experiment constituted test day 2. On this day, animals were treated in a manner identical to that employed on test day 1.

Blood Ethanol Determinations

Forty-eight additional animals were used to determine whether taurine influenced the overall metabolism of ethanol at the intermediate dose of ethanol used in this study. Animals were injected with 1.2 g/kg ethanol and taurine (0.0 and 45 mg/kg). These animals were then sacrificed by decapitation at 15, 30, and 60 min postinjection. Trunk blood was collected and later assayed for ethanol levels by head space gas chromatography with a flame-ionization detector (11).

RESULTS

In line with the consensus in the field of CTA, saccharin intake data is expressed as percent change from baseline intake. There were no differences in the ratio of saccharin intake on pairing day 1 to mean water intake on the day prior to that across all groups. Baseline constituted the saccharin intake scores on pairing day 1. Figure 1 illustrates the effects of taurine on the CTAs induced by several doses of ethanol. A three-way analysis of variance (ANOVA) with repeated measures on the last factor (taurine × ethanol × testing days)

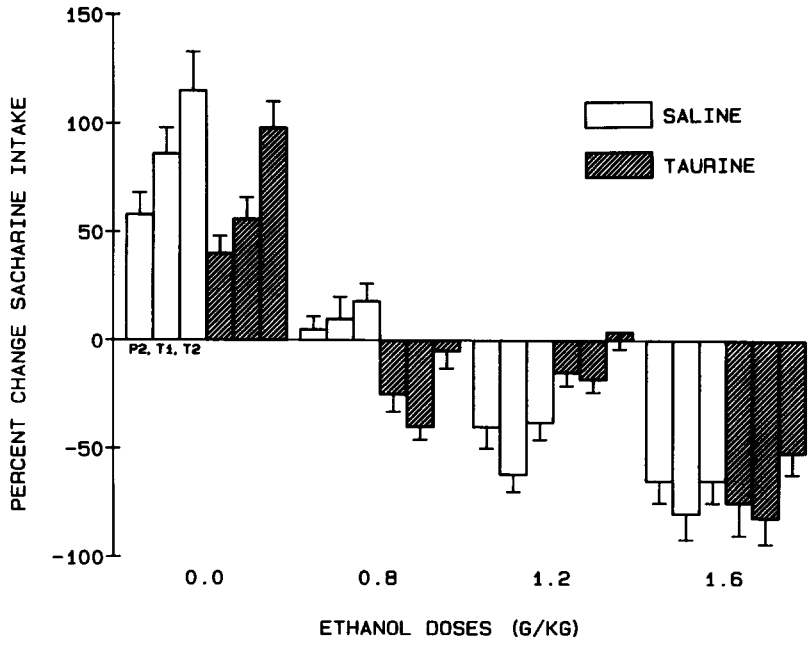

FIG. 1. Effect of taurine (45 mg/kg) on ethanol-induced conditioned taste aversion. Bars represent mean ± SEM percentage change in the intake of saccharin on test days ($P_2T_1T_2$) from that on pairing day 1 for animals treated with taurine or saline and ethanol (0.8, 1.2, and 1.6 g/kg).

was conducted on mean percent change from baseline scores of saccharin intake. Table 1 summarizes the data analysis following the application of the three-way ANOVA. Specific comparisons using Student's *t*-test were made using marginal means. From these comparisons, one can see that taurine interacted with ethanol in a biphasic manner dependent upon the doses of ethanol. Thus, at 0.8 g/kg ethanol the taurine treatment significantly enhanced the effects of ethanol to result in a significant CTA ($p < 0.05$). At the intermediate ethanol dose of 1.2 g/kg, taurine significantly attenuated the CTA induced by ethanol ($p < 0.05$). At the high ethanol dose of 1.6 g/kg, taurine had no significant effect on the CTA induced by that dose ($p > 0.05$).

Table 2 summarizes the data obtained on the effects of taurine on blood ethanol levels following administration of the intermediate dose of ethanol used in the experiment. As can be seen from the table, taurine was not significantly different from saline in their effects on blood ethanol levels at three time intervals following ethanol administration ($p > 0.05$).

TABLE 1

SUMMARY TABLE OF THREE-WAY (ANOVA) WITH REPEATED MEASURES ON THE LAST FACTOR (TAURINE × ETHANOL × TESTING DAYS)

Source of Variation	*df*	*F*	*p*
Taurine (T)	1	0.574	0.4514
Ethanol (E)	3	59.472	0.0000
T × E	3	5.303	0.0025
Error	66		
Testing days (TD)	2	28.098	0.0000
T × TD	2	0.006	0.9937
E × TD	6	3.689	0.0020
T × E × TD	6	0.665	0.6782
Error	132		

DISCUSSION

The results obtained in the present study demonstrate that taurine by itself does not induce a CTA (see Fig. 1). On the other hand, the results show a significant interaction between taurine and ethanol in their effects on ethanol-induced CTA in rats. This interaction was biphasic in nature and dependent upon the specific dose of ethanol used. At the lowest ethanol dose (0.8 g/kg), which in itself resulted in a marginal CTA, taurine significantly enhanced the CTA induced by this dose. The intermediate ethanol dose of 1.2 g/kg produced a significant CTA. This CTA was blocked by administration of taurine. Finally, the CTA produced by the high dose of ethanol

TABLE 2

EFFECT OF TAURINE (0 AND 45 mg/kg) ON MEAN (mg/kg) PLASMA ETHANOL LEVELS ± SEM AT DIFFERENT TIMES AFTER ETHANOL ADMINISTRATION (1.2 g/kg)

	Time (min)		
	15	30	60
Saline	1.46 ± 0.11	1.72 ± 0.14	1.16 ± 0.04
Taurine	1.36 ± 0.12	1.64 ± 0.12	1.10 ± 0.08

$n = 8$ per group.

(1.6 g/kg) was not affected by administration of taurine. The dose of taurine (45 mg/kg) used in this the present study was chosen on the basis of results from several studies (3,4,10) demonstrating that it was the optimal dose in effecting behavior. These data are similar to those obtained in an earlier study (3) reported by us on the effects of taurine on ethanol-induced motor activity in mice. The data obtained in this study together with our earlier study (3) support the findings reported by other investigators that taurine exerts a significant effect on ethanol-induced behaviors (4,10,13). Further, the present study supports our findings that the interaction between taurine and ethanol is biphasic and dependent upon the doses of ethanol (3) and the effects of taurine on ethanol-induced behaviors are not restricted only to the narcotic effects of ethanol and in fact extend over a wide range of ethanol-induced changes in behavior. In this context, it is important to note that the effects of taurine in this study could not be ascribed to an effect of taurine on peripheral ethanol metabolism. The data obtained in this study shows that blood ethanol levels measured up to 60 min following its administration were not affected by administration of taurine. This finding within the context of the other studies from this and other laboratories (3,4,10,13) suggests that the effects of taurine on ethanol-induced behaviors are mediated centrally and not peripherally.

While at present the precise mechanism underlying the interaction between taurine and ethanol remains unclear, the above suggestion is somewhat at variance with a report by Watanabe et al. (15), who (16) observed that taurine significantly reduced the rise in blood and liver acetaldehyde that normally followed ethanol loading in rats. These authors (15) also reported that an increase in blood acetaldehyde and not ethanol following ethanol ingestion was also reduced by administration of taurine. Watanabe et al. (16,17) suggested that these effects of taurine may be a result of the activation of hepatic aldehyde dehydrogenase by taurine. Given the large body of evidence implicating acetaldehyde in the mediation of many of the psychopharmacological properties of ethanol (1,2) together with evidence that inhibition of brain aldehyde dehydrogenase activity modified ethanol-induced CTA (14), it is suggested that this effect of taurine on the formation of acetaldehyde through activation of aldehyde dehydrogenase could be the mechanism underlying the results obtained in the present study. However, one must note that this explanation still does not completely account for the facilitation of CTA observed with taurine-low-dose ethanol administration. Several experiments are now in progress in our laboratory to examine this possibility.

ACKNOWLEDGEMENTS

This study was in part supported by a grant from FCAR of Canada to Z.A., B.R.S., and C.M.G.A.

REFERENCES

1. Aragon, C. M. G.; Abitbol, M.; Amit, Z. Ethanol-induced CTA mediated by acetaldehyde through central catecholamine activity. Psychopharmacology (Berl.) 103:74–77; 1990.
2. Aragon, C. M. G.; Spivak, K.; Amit, Z. Blockade of ethanol induced conditioned taste aversion by 3-amino-1,2,4,-triazole: Evidence for catalase mediated synthesis of acetaldehyde in rat brain. Life Sci. 37:2077–2084; 1985.
3. Aragon, C. M. G.; Trudeau, L.-E.; Amit, Z. Effect of taurine on ethanol-induced changes in open-field locomotor activity. Psychopharmacology (Berl.) 107:337–340; 1992.
4. Boggan, W. O.; Medberry, C.; Hopkins, D. H. Effects of taurine on some pharmacological properties of ethanol. Pharmacol. Biochem. Behav. 9:469–472; 1978.
5. Bures, J.; Buresova, O. Physiological mechanisms of conditioned food aversion. In: Milgram, N. W.; Krames, L.; Alloway, T. M., eds. Food aversion learning. New York: Plenum Press; 1977:219–255.
6. Cappell, H.; LeBlanc, A. E.; Endreryi, L. Aversive conditioning by psychoactive drugs: Effects of morphine, alcohol and chlordiazepoxide. Psychopharmacologia 29:239–246; 1973.
7. Chapman, G. E.; Greenwood, C. E. Taurine in nutrition and development. Nutr. Res. 8:955–968; 1988.
8. Hanretta, A. T.; Lombardini, J. B. Is taurine a hypothalamic neurotransmitter? Brain Res. Rev. 12:167–201; 1987.
9. Huxable, R. J.; Bressler, R. Effect of taurine on a muscle intracellular membrane. Biochem. Biophys. Acta 323:575–583; 1973.
10. Iida, S.; Hikichi, M. Effect of taurine on ethanol-induced sleeping-time in mice. J. Stud. Alcohol 37:19–26; 1976.
11. Iversen, H. L.; Damgaard, S. E. Determination of acetaldehyde in human blood using thiourea to inhibit ethanol interference. Clin. Chim. Acta 135:151–158; 1983.
12. Kaczmarek, L.K.; Davison, A. N. Uptake and release of taurine from rat brain slices. J. Neurochem. 19:2355–2362; 1972.
13. McBroom, M. J.; Elkhawad, A. O.; Dlouha, H. Taurine and ethanol-induced sleeping time in mice: Route and time course effects. Gen. Pharmacol. 17:97–100; 1986.
14. Spivak, K.; Aragon, C. M. G.; Amit, Z. Alterations in brain aldehyde dehydrogenase activity modify ethanol-induced conditioned taste aversion. Alcohol. Clin. Exp. Res. 11:513–517; 1987.
15. Watanabe, A.; Hobara, N.; Kobayashi, M.; Nagashima, H. Effect of taurine on blood acetaldehyde elevation following alcohol ingestion. Res. Comm. Subst. Abuse 6:247–250; 1985.
16. Watanabe, A.; Hobara, N.; Nagashima, H. Lowering of liver acetaldehyde but not ethanol concentrations by pretreatment with taurine in ethanol-loaded rats. Experientia 41:1421–1422; 1985.
17. Watanabe, A.; Hobara, N.; Nagashima, H. Activation and inhibition of yeast aldehyde dehydrogenase activity by pantethine and its metabolites. Ann. Nutr. Metab. 30:54–57; 1986.
18. Wu, J.-Y.; Lin, C.-T.; Thalmann, R.; Taber, K.; Song, G. X.; Su, Y. Y. T. Immunocytochemical and physiological identification of taurine neurons in the mammalian CNS. In: Oja, S. S.; Ahtee, L.; Kontro, P.; Paasonen, M. K., eds. Taurine: Biological actions and clinical perspectives. New York: Alan R. Liss; 1985: 261–270.