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Effect of Taurine on Some Pharmacological Properties of Ethanol[1]

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W. O. BOGGAN, C. MEDBERRY AND D. H. HOPKINS. *Effect of taurine on some pharmacological properties of ethanol.* PHARMAC. BIOCHEM. BEHAV. 9(4) 469–472, 1978.—Taurine was effective in reducing the hypnotic effect of ethanol but did not antagonize the effect of ethanol on seizure susceptibility, body temperature, or brain 5-HIAA concentration.

Taurine Pharmacological effects Ethanol

IT HAS BEEN reported [17] that acute, simultaneous injection of taurine (2-aminoethanesulfonic acid) reduces ethanol induced sleep time of mice. This reduction was substantially greater than that produced by 9 other amino acids which were administered at a similar concentration. Several possible mechanisms may underlie this antagonism. For example, it has been postulated that taurine is a neuromodulator or inhibitory transmitter. In support of these hypotheses are the following: iontophoretically administered taurine causes a depressor effect on most neurons [5, 6, 12, 14]; taurine appears to be concentrated in synaptosomal fractions [7,29] (though it is possible that it occurs within glia cells [9]); it can be released by chemical and electrical stimulation [18] and be taken up into brain slices [20].

Taurine may be a membrane stabilizer on the basis of data from the sacroplasmic reticulum from rat skeletal muscle wherein taurine caused an increase in the rate of calcium oxalate uptake, an increase in the total calcium sequestered, and a decrease in the rate of calcium transport after treatment of the preparation with phospholipase C [16]. An increase in the mitochondrial binding of calcium after taurine has also been reported [8]. Furthermore, taurine in the intact dog heart prevents the efflux of potassium which accompanies large doses of epinephrine [25]. Much of this work has been summarized [3,4].

A third possible mechanism for the reduction of ethanol induced sleep time by taurine was suggested by Iida and Hikichi [17]. They speculated that taurine may form a complex with ethanol or acetaldehyde or increase the metabolic rate of ethanol. One purpose of our investigation was to examine the effect of taurine on circulating levels of ethanol.

Since taurine did antagonize the effects of ethanol on sleep time [17] we questioned whether taurine would also antagonize some of the other reported effects of ethanol. Specifically, we attempted to replicate the effect of taurine on ethanol induced sleeping time and to study its effects on ethanol induced decreases in body temperature [11, 26, 32], seizure susceptibility [1, 23, 36], and increase in brain 5-hydroxyindole acetic acid [31].

METHOD

Adult male C57BL/6J mice purchased from the Jackson Laboratories (Bar Harbor, Maine) were used in these studies. All animals were housed 5 per cage in a 7:00 p.m.–7:00 a.m. dark-light cycle with continuous access to water and Wayne Mouse Breeder Blox. The mice were randomly assigned to the experimental groups and each animal was used only once. In every experiment, each taurine solution was adjusted to pH 7.0 with dilute NaOH.

Taurine and Ethanol Induced Sleeping Time

Mice were fasted overnight with continuous access to H_2O. At 10 a.m. the next morning they were injected intraperitoneally with ethanol (3.5 g/kg, 0.2 ml/10 g body wt. prepared from 95% ethanol) or with ethanol plus taurine (45 mg/kg; 0.2 ml/10 g body wt.). Sleeping time was measured as the total elapsed time from the initial loss of righting reflex to the animals' return without remission to all 4 feet. The righting reflex was considered lost if the animal, when placed on its back, did not right itself. The sleep time experiment was conducted in a constant temperature room 25 ± 0.1°C and was repeated 3 times. The data were analyzed by *t*-tests.

Taurine and Ethanol Induced Hypothermia

Baseline rectal temperatures of mice were taken by inserting a small animal probe attached to a telethermometer (Yellow Springs Instrument, Model 43TA) approximately 3 cm into the colon. The animals were lightly restrained by the tail during this procedure. Initial mean rectal temperatures of the groups were determined to be equal prior to the onset of the experiments. The mice were then injected with ethanol (2 g/kg; 0.2 ml/10 g body wt.) or with ethanol plus taurine (30

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mg/kg; 0.2 ml/10 g body wt.; this dose of taurine was chosen for this experiment because higher doses were found to produce hypothermia by themselves). The temperature was taken again 30, 60, 120 and 240 min after injection. These studies were carried out in a constant temperature room of 25 ± 0.1°C and the data were analyzed by analysis of variance on repeated measures.

Taurine and Ethanol Antagonism of Electrically Induced Seizures

Mice were injected intraperitoneally with ethanol (3 g/kg), ethanol plus taurine (45 mg/kg), H_2O, or taurine 45 mg/kg. The volume of all injections was 0.2 ml/10 g body wt. The animals were tested one half hour later for susceptibility to electrically induced seizures (ECS). ECS was administered transcorneally by an apparatus [13] which consisted of an 800 V transformer with a fixed resistor in series with the mouse to produce a current of 12 mA r.m.s. An electromechanical timer was used to obtain an ECS duration of 200 msec. Incidence of muscle spasms, clonic, tonic seizures and death were recorded. These data were analyzed by use of Chi Square.

Taurine and Blood Ethanol Concentration

Mice were injected intraperitoneally with ethanol (3.5 g/kg, 0.2 ml/10 g body wt.) or with ethanol plus taurine (45 mg/kg; 0.2 ml/10 g). The blood concentration of ethanol was measured by the method of Lundquist [22] 30, 60, 90, and 120 min later. These data were analyzed by analysis of variance on repeated measures.

Taurine and Ethanol on Brain 5-hydroxyindole acetic acid (5-HIAA) and 5-HT

Four groups of animals were used in this experiment: H_2O, taurine, ETOH, and ETOH plus taurine. The animals (N=8/group) were injected with taurine (45 mg/kg) or its vehicle and then 1 hr later with ETOH (3 g/kg) or its vehicle. They were sacrificed 2 hr after the last injection. Two hr has been previously shown to be the optimal time to see the increase in brain 5-HIAA after ETOH [31]. The brains were analyzed for 5-hydroxytryptamine (5-HT) and 5-HIAA by the method of Jonsson and Lewander [19]. These data were analyzed by analysis of variance and *t*-tests.

RESULTS

Taurine significantly reduced the sleeping time of animals receiving ethanol. Ethanol (N=23)=47.3 ± 5.5 min; Ethanol plus Taurine (N=23)=25.9 ± 3.6 min, $t=3.18$, $p<0.005$. Data are given as mean ± S.E.

Although ethanol administration decreased body temperature as a function of time $F(2,28)=63.25$, $p<0.01$, taurine did not antagonize these effects (Table 1). Taurine also failed to antagonize the effects of ethanol on electroconvulsive shock (Table 2) and to alter the amount of circulating ethanol (Table 1).

Ethanol produced a significant ($p<0.05$) increase in brain 5-HIAA (447.7 ± 16.4) as compared to its control (383.8 ± 14.6). This effect was not antagonized by giving taurine before the ethanol (439.5 ± 31.1) nor did taurine produce any effect on 5-HIAA by itself (371.6 ± 9.0). The values are given as ng/g ± S.E. No differences in 5-HT concentration were found between the groups.

DISCUSSION

In agreement with the study of Iida and Hikichi [17] we have found that taurine significantly reduces the sleep time of C57BL/6J mice when administered simultaneously with ethanol. This effect does not seem to be due to differential rates of absorption of ethanol from the blood stream since no differences in circulating ethanol were observed between animals treated with ethanol and those treated with taurine plus ethanol.

The same dose of taurine (45 mg/kg) given at the same time as the study on sleep time was ineffective in altering the effects of alcohol on seizure susceptibility and did not exhibit anticonvulsant properties by itself. Previous studies have at-

TABLE 1

EFFECT OF TAURINE ON ETHANOL INDUCED CHANGES IN TEMPERATURE AND BLOOD ETHANOL CHANGES

	Time (Min) N		0	30	60	90	120	240
					Temperature			
ETOH	8	Mean ±	37.8	35.4	35.9	—	36.9	36.9
		S.E.	0.362	0.159	0.262		0.085	0.279
ETOH + Taurine*	8	Mean ±	37.2	35.7	35.6	—	36.6	37.2
		S.E.	0.533	0.31	0.289		0.210	0.585
					Blood Alcohol (mg/100 ml)			
ETOH	7	Mean ±	—	362	317	320	283	
		S.E.		023	031	013	021	
ETOH + Taurine*	7	Mean ±	—	363	341	336	291	
		S.E.		026	028	018	011	

*Taurine 30 mg/kg, ETOH 2 g/kg given together.
**Taurine 45 mg/kg, ETOH 3.5 g/kg given together.

TABLE 2

EFFECT OF TAURINE ON ETHANOL INDUCED DECREASE IN SEIZURE SUSCEPTIBILITY*

Group	N	Number Responding			
		MS‡	Clonic	Tonic	Death
H_2O	15	15	15	10	7
Taurine	15	15	15	11	8
ETOH	10	0	0	0	0
ETOH+Taurine†	10	0	0	0	0

*ECS given transcorneally at 12 mA for 0.2 sec.
†ETOH=3 g/kg; Taurine=45 mg/kg; Drugs given together.
‡MS=muscle spasms.

tributed to taurine the capacity to block the epilepsy produced by acute and chronic cobalt lesions in mice [34], to possess anticonvulsant properties when applied as pretreatment to mice subsequently undergoing "standard" anticonvulsant testing under conditions of osmotic stress [33] and to block audiogenic seizures in rats when given intracerebrally [21]. Emson [10], using the cobalt-induced epileptic focus in the rat, was unable to show any significant anticonvulsant action of taurine. Explanations for the lack of effectiveness of taurine as an anticonvulsant in our study as well as that of Emson [15] are not available at this time.

Ethanol induced hypothermia is well documented [11, 26, 32] and supported by our findings. Taurine at a concentration effective in reducing ethanol induced sleep time by 45% did not modify the hypothermic effect of ethanol or produce a hypothermia itself. Our pilot studies at a higher dose of taurine and other studies have reported that taurine would produce hypothermia in mice [15] and rats [28].

Acute ethanol administration has been associated with an increase in the levels of the major acid metabolite of serotonin, 5-hydroxyindole-3-acetic acid (5-HIAA) in mice [31]. This increase was found to be due to inhibition of transport of 5-HIAA from the brain via the choroid plexis [32]. Examination of this phenomenon with respect to taurine and taurine plus ethanol should not only allow us to evaluate the effect of low doses of taurine upon serotonin (5-HT) and 5-HIAA levels, but would also permit determination of any modifications elicited by taurine on ethanol produced alterations in serotonin metabolism. Taurine administration prior to ethanol neither modified the ethanol mediated increase in 5-HIAA, nor significantly affected either 5-HT or 5-HIAA levels.

It is indeed possible that higher concentrations of taurine might protect against the pharmacological effects of ETOH not modified in the present experiments. It is of interest, however, that taurine at the present dosage will modify the hypnotic action of alcohol and not the others. It seems unreasonable to suggest that sleep time is just a more sensitive measure than temperature, or seizure susceptibility, or brain 5-HIAA concentration, especially since the sleep time in experimental animals was 45% that of controls. Since those mechanisms controlling sleep time are probably different from those controlling seizure susceptibility and temperature regulation taurine must have a selective mode of action able to modify that mechanism, but not the others.

Given that taurine does antagonize at least ethanol induced sleep time, questions must be asked as to how this might happen. One possible mechanism for taurine to antagonize the action of alcohol is by stabilization of tissue calcium. Ross [27] has reported that ethanol and salsolinol acutely administered to rats creates a dose dependent decrease in brain calcium levels. Taurine has been shown to be able to stabilize calcium levels in the brains of rats which have been subjected to altered electrolyte status by injection of glucose [35]. Whether this is happening in mice and whether calcium plays a role in ethanol induced sleep is not known.

A second possible mechanism by which taurine might antagonize the effects of ethanol is by complexing with acetaldehyde, the metabolic product of ethanol. Salsolinol, the Pictet-Spengler condensation product of dopamine and acetaldehyde and tetrahydro-β-carboline, the serotonin derivative of the same condensation reaction with acetaldehyde, have been proposed as the major components in the CNS etiology of ethanol [24]. Current studies by Alivasatos *et al.* [2] concerning the formation of thiazolidine compounds by the reaction of substituted aldehydes and the sulphur moiety of cysteine propose that cysteine arrests the reactants of the Pictet-Spengler condensation. Sprince and coworkers [30] have examined the effect of several sulfur containing compounds including taurine and its noncarbohydrate analog sodium metabisulfite on acetaldehyde induced anesthesia (loss of righting reflex) and lethality. Pretreatment with the test compound, orally, 30–45 min before acetaldehyde suggested that N-acetyl-L-cysteine, thiamine HCl, sodium metabisulfite, and L-cysteic acid at a dosage of 2 mM/kg were effective protectants against loss of righting reflex and death. Taurine was found to be one of the least effective protectants; however, the dosages used were rather low.

It is recognized that the studies reported herein are limited because of the lack of dose response curves for both taurine and ethanol; however the data do provide new information about the interaction of these 2 compounds. Though the potential of taurine as a broad spectrum antagonist of ethanol is in doubt because of these data, it does perhaps provide a tool and guide for examining one aspect of ethanol action (hypnosis).

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