Effect of ethanol on taurine concentration in the brain[1]

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Summary. The taurine concentration in the brain was decreased in ethanol-dependent rats, but returned to normal soon after withdrawal of ethanol. It was not affected by acute ethanol administration.

Taurine is present in high concentration in the central nervous system and has many of the characteristics of an inhibitory neurotransmitter or neuromodulator[2-6]. Previously, we showed that this amino acid in the brain may be related to the development of barbiturate-dependence, or the appearance of withdrawal signs[7]. Recently, Iida and Hikichi[8] and Boggan et al.[9] reported that taurine was effective in reducing the hypnotic effect of ethanol in mice. Furthermore, Ikeda[10] showed that taurine had a beneficial effect in preventing and controlling withdrawal symptoms in chronic alcoholics. These findings suggest that the effects of acute and chronic administration of ethanol may involve taurine metabolism in the brain. We examined the effects of acute and chronic ethanol administration and ethanol withdrawal on the taurine concentration in rat brain to obtain information about the involvement of brain taurine in the action of ethanol. We also reexamined the effect of taurine administration on the ethanol-induced sleeping time.

Materials and methods. Male Sprague-Dawley rats (200 to 240 g) were rendered ethanol-dependent by the method of Majchrowicz[11], which consists of administering doses of 11-15 g/kg daily by intragastric intubation in 3-5 fractions over a period of 4 days. One third of the dependent rats showed spontaneous convulsions after withdrawal of ethanol. For studying the effect of an acute dose of ethanol, male Sprague-Dawley rats (120 g) were starved overnight before the experiment. Rats were decapitated at a definite time of day (09.00-11.00 or 22.00 h) to avoid the effect of diurnal variation in the brain taurine concentration[12]. The brain was removed rapidly and cut into 3 parts (cerebral cortex, brain stem and cerebellum), and taurine was determined as described previously[7,12]. Blood ethanol was determined by the enzymatic method of Jones et al.[13]. In studies on the sleeping time, experiments were done between 10.00 and 13.00 h at a constant temperature (22±1 °C), using male *dd* and *JCL/ICR* mice. The sleeping time was measured as the total time from the initial loss of the righting reflex to the animal's return without remission. Mice which did not sleep were omitted from the experiment.

Results and discussion. Table 1 shows the taurine concentrations in the cerebral cortex, brain stem and cerebellum of ethanol-treated rats. Acute administration of ethanol did not affect the taurine concentration in any of these regions, in agreement with previous results of Häkkinen et al.[14] and Flock et al.[15]. The ethanol-dependent rats used in this study exhibited withdrawal signs, such as tail stiffness, stereotypic head movements, hyperirritability, tremors, jumping and convulsions, once blood ethanol had decreased (usually 12-15 h after the last administration). The blood ethanol concentrations of ethanol-dependent rats at 1, 6, 12 and 18 h after the final dose were 329±28, 171±31, 57±16 and 28±7 mg/dl, respectively (n=10-12). The taurine concentration in all regions examined was significantly lower in ethanol-dependent rats than in controls, but the concentrations returned to normal after ethanol withdrawal. After withdrawal, the taurine concentration increased particularly rapidly in the cerebral

Table 1. Effects of acute and chronic administration of ethanol and of ethanol withdrawal on brain taurine concentration in rats

Treatment		Hours after withdrawal	Taurine concentration (μmole/g wet weight)		
			Cerebral cortex	Brain stem	Cerebellum
Acute	0 g/kg, p.o., 1h		6.0±0.4	4.6±0.3	5.5±0.4
	2.5		5.5±0.7	4.1±0.1	5.4±0.3
	5.0		6.5±0.2	4.4±0.2	5.9±0.2
Chronic	Control		6.9±0.3	5.5±0.1	6.8±0.2
	Dependent	0	5.3±0.2[b]	4.7±0.1[b]	5.8±0.1[a]
		12	8.0±0.5[d]	5.1±0.2	6.2±0.2
		24	6.3±0.3[c]	5.4±0.3	6.6±0.3
		72	6.4±0.2[c]	5.6±0.2[d]	6.8±0.1[d]

[a] $p<0.05$, [b] $p<0.01$, compared with control value; [c] $p<0.05$; [d] $p<0.01$ compared with value at 0 time after ethanol withdrawal.

Table 2. Effect of taurine on ethanol-induced sleeping time in mice

Strain	Ethanol (g/kg)	Taurine (mg/kg)	N	Sleep onset (sec)	Sleep duration (sec)
JCL/ICR	3.5	0	8	124± 9	390± 48
		50	5	130±35	498± 53
		100	7	106± 9	397± 64
dd	4.0	0	27	107± 5	1708±188
		30	20	115± 4	1437±165
		50	17	111± 7	1753±238

Taurine was injected i.p. 5 min before ethanol administration (20 (w/v)%, i.p.).

cortex, parallel with the appearance of withdrawal signs. We also tried to produce physical dependence by giving increasing concentration of ethanol (2.5–17 g/kg daily) in the drinking water over a period of 8 weeks. But, these rats did not show apparent withdrawal signs. Moreover, the taurine concentration in their cerebral cortex did not change, though they showed slightly decreased levels of taurine in the brain stem and cerebellum. These findings suggest that taurine in the cerebral cortex may be involved in development of ethanol dependence or the appearance of withdrawal signs. Administration of taurine (0.6 g/kg, p.o., daily during ethanol administration) did not affect withdrawal signs of ethanol-dependent rats. It is unlikely that this negative result was due to poor incorporation of injected taurine into the brain, since taurine administration prevented decrease in the taurine concentration of ethanol-dependent rats (data not shown). Thus, it is still uncertain how changes in brain taurine are related to the appearance of withdrawal signs.

Table 2 shows the effect of taurine on the ethanol-induced sleeping time. In contrast to previous reports[8,9], taurine did not affect the sleeping time in either *JCL/ICR* or *dd* strain mice. We used 2 different strains, because it is well known that there are strain differences in the hypnotic action of ethanol[16,17]. Iida and Hikichi[8] first reported the antagonistic effect of taurine on the ethanol-induced sleeping time in *dd* mice. This effect was also observed by Boggan et al.[9] in C57BL/6J mice. But it is unlikely that the injected taurine entered the brain in these experiments, since exogeous taurine is incorporated into the brain only very slowly[18–21]. We observed that the taurine concentration in the brain was not changed by taurine administration under the same experimental conditions. Furthermore, we found that administration of taurine did not affect the ethanol concentration in the blood or brain of ethanol-treated mice (data not shown), indicating that taurine administration does not affect the metabolism of ethanol. Thus, at present, we cannot explain why our results differ from those of previous authors.

The present findings suggest that in the brain taurine, like some other putative neurotransmitters[23], is involved in the effect of chronic, but not acute, ethanol administration.

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Effects of probenecid on plasma/tissue distribution of ^{14}C-benzylpenicillin in rats

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Summary. Probenecid (50 mg·kg^{-1}) was found to induce an increase of the plasma concentration of ^{14}C-benzylpenicillin with a decrease of the concentration in liver and kidney. Accumulation in corresponding tissue slices was reduced by probenecid. Therefore, the well known increase of penicillin in plasma after probenecid seems to be not only due to an inhibition of renal excretion but also to a reduced tissue uptake in liver and kidney.

Probenecid, long known as an inhibitor of the tubular transport of weak organic anions[1–3], was widely used in therapy with penicillin to yield higher plasma levels. Numerous reports on the efficacy of this therapy and on probenecid combination with other antibiotics are present[4–6]. Today, at least in the case of certain strains of gonococci which are relatively resistent to penicillin G, ampicillin or amoxycillin, the combination with probenecid is indicated[7]. Nevertheless, the interactions of probenecid and penicillin are not completely understood. In some experimental studies extrarenal effects of probenecid have been demonstrated[8,9] but there is no reference to penicillin concentration in the tissue after probenecid. Therefore we studied in vitro and in vivo the plasma/tissue distribution of ^{14}C-benzylpenicillin under the influence of probenecid.

Methods. Male albino rats (NMRI - Hannover) weighing 360±60 g obtained 25 mg equivalent to 41,700 IE ^{14}C-benzylpenicillin/kg b.wt i.p. The labelled (Amersham Corp.) and the non-labelled benzylpenicillin (pharmacy, AK Rissen) were mixed to yield an appropriate sp. act. of