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Ethanol markedly increases ''GABAergic'' neurosteroids in alcohol-preferring rats

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Abstract

Alcohol administration (1 g/kg, i.p.) increased the levels of the neurosteroids 3α-hydroxy-5α-pregnan-20-one (allopregnanolone) and 3α,21-dihydroxy-5α-pregnan-20-one (allotetrahydrodeoxycorticosterone, THDOC) in the cerebral cortex and hippocampus both in alcohol-naive Sardinian alcohol-preferring (sP) and -non-preferring (sNP) rats (two rat lines selectively bred for alcohol preference and non-preference, respectively). However, the increase reached several fold higher levels in sP than in sNP rats (6–24 vs. 2–11 fold the basal levels, respectively). Since the two neurosteroids are the most potent endogenous positive modulators of $GABA_A$ receptors and elicit anxiolytic and rewarding effects, while voluntary alcohol consumption produces anxiolytic and rewarding effects in sP but not in sNP rats, the results suggest that the neurosteroids may play a role in the anxiolytic and rewarding effects of alcohol in sP rats.

Keywords: Brain neurosteroid; Alcohol; Sardinian alcohol-preferring (sP) and -non-preferring (sNP) rat

The endogenous neurosteroids 3α-hydroxy-5α-pregnan-20-one (allopregnanolone, AP) and 3α,21-dihydroxy-5α-pregnan-20-one (allotetrahydrodeoxycorticosterone, THDOC) are the most potent endogenous positive modulators of $GABA_A$ receptors and therefore they exert sedative, hypnotic, anticonvulsant and anxiolytic effects (see Lambert et al., 1995). There is evidence for interactions between alcohol and the neurosteroids at $GABA_A$ receptors (see Grobin et al., 1998). Alcohol too positively modulates $GABA_A$ receptors and exerts sedative and anxiolytic effects. Allopregnanolone and THDOC completely substitute for the discriminative stimulus effects of alcohol both in rats and monkeys (see Grobin et al., 1998). Neurosteroids have been shown to be rewarding in rodents (Finn et al., 1997).

The aim of the present study was to elucidate if alcohol administration would modify neurosteroid concentrations in the rat brain. In order to correlate changes in neurosteroids with the anxiolytic and rewarding properties of alcohol, the effects of the drug on brain neurosteroids were studied in Sardinian alcohol-preferring (sP) and -non-preferring (sNP) rats, two rat lines selectively bred for alcohol preference and non-preference, respectively (see Colombo, 1997). Moreover, sP rats exhibit a genetically determined high level of anxiety-related behaviors, which are suppressed by voluntary alcohol consumption (see Colombo, 1997). In addition, alcohol exhibits rewarding effects in sP but not in sNP rats, as demonstrated by induction of conditioned place preference solely in the former rats (Ciccocioppo et al., submitted).

Male ethanol-naive sP ($n = 14$) and sNP ($n = 14$) rats, from the 42nd generation and 3 months old, were used. Rats were housed individually under standard environmental conditions. On the test day, rats were injected intraperitoneally with a dose of alcohol (1 g/kg) in the range of the amounts that sP rats would voluntarily consume in binges during the nocturnal phase of the light/dark cycle (see Colombo, 1997). Brain and plasma steroid extraction and measurement were carried out as previously described

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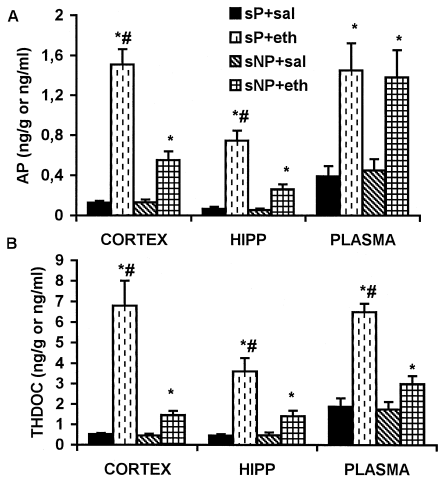

Fig. 1. Effect of the acute, intraperitoneal administration of 1 g/kg alcohol on the levels of 3α-hydroxy-5α-pregnan-20-one [allopregnanolone (AP), panel A] and 3α,21-dihydroxy-5α-pregnan-20-one [allotetrahydrodeoxycorticosterone (THDOC) panel B] in cortex, hippocampus and plasma of sP and sNP rats. AP and THDOC levels in the two brain areas and plasma were expressed in ng/g and ng/ml, respectively. Each bar is the mean ± SEM of seven subjects. $^{*}P < 0.05$ with respect to saline-treated rats of the same line; #: $P < 0.05$ with respect to ethanol-treated sNP rats.

(Barbaccia et al., 1997). Data were analyzed by one-way ANOVA followed by the two-tailed Student's *t*-test for post-hoc comparison.

As the Fig. 1 shows, alcohol increased allopregnanolone and THDOC concentrations in the cerebral cortex and hippocampus in both rat lines, but the increase was several fold higher in sP than in sNP rats (AP: 17 vs. 5 in cortex and 11 vs. 4 in hippocampus of sP and sNP rats, respectively; THDOC: 24 vs. 4 in cortex and 6 vs. 2 in hippocampus of sP and sNP rats, respectively). Plasma concentrations of allopregnanolone were similarly enhanced by alcohol in both rat lines, but plasma concentrations of THDOC increased to a greater extent in sP than in sNP rats, suggesting that alcohol stimulates more efficiently the neurosteroidogenesis in sP rats, not only in brain but in peripheral organs as well.

Brain allopregnanolone and THDOC concentrations have been shown to be increased by different stressors; the increase has been interpreted as a compensatory mechanism to restore GABAergic transmission reduced by stress (Barbaccia et al., 1997). In fact, exogenously administered allopregnanolone and THDOC attenuate stress-induced release of CRF, ACTH and corticosterone and blunt the anxiogenic effect of CRF. Since alcohol is known to be a stressor and to activate hypothalamus–pituitary–adrenal axis, the finding that it increases brain neurosteroid concentrations was expected from such premises. However, the finding that a small dose of alcohol, as that voluntarily consumed by sP rats, produces such a remarkable increase in GABAergic neurosteroids in the sP rat brain suggests that allopregnanolone and THDOC not only counteract the stressful effects of alcohol but do also disclose the anxiolytic and rewarding properties of this drug, and therefore play an important role in alcohol preference.

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