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BRIEF COMMUNICATION

Decreased Intoxicating Effect of Ethanol in Rats After 6-Hydroxydopamine-Induced Degeneration of Ascending Dopamine Pathways

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KIIANMAA, K. *Decreased intoxicating effect of ethanol in rats after 6-hydroxydopamine-induced degeneration of ascending dopamine pathways.* PHARM. BIOCHEM. BEHAV. 9(3) 391–393, 1978.—Selective lesion of ascending dopamine pathways was made by injecting the neurotoxin 6-OHDA (8 μg/4 μl) bilaterally close to the nigro-striatal dopamine pathway of 18 male Long Evans rats. Similar injections of the vehicle were given to 10 control rats. Two months after the operation intoxication was measured in a tilting-plane test after an injection of ethanol (2 g/kg, IP). Ethanol impaired the performance of the 6-OHDA-treated rats significantly less than that of the controls. This finding suggests a role for the central dopamine neurons in the intoxicating effect of ethanol.

Alcohol Ethanol intoxication Dopamine 6-Hydroxydopamine

ONE of the most conspicuous effects of ethanol is the impairment of motor coordination. Very little work has, however, been done to clarify which neuronal systems are involved in this aspect of ethanol intoxication. Although cholinergic systems are involved in motor functions, a non-cholinergic basis and non-cortical locus for alcohol-induced impairment was suggested by the finding that ethanol caused motor deficits more or less independently of cortical influences in rats [16]. In addition to acetylcholine, the central dopamine systems are also known to be involved in motor functions [13]. In order to clarify the role of the central dopamine neurons in ethanol intoxication, the neurotoxin 6-hydroxydopamine (6-OHDA) was used in the present study to damage 2 ascending dopamine pathways, one innervating the corpus striatum and the globus pallidus (the nigro-striatal dopamine system) and the other innervating the nucleus accumbens and the tuberculum olfactorium (the meso-limbic dopamine system) [20].

MATERIALS AND METHOD

Male Long Evans hooded rats, 3 months old at the beginning of the experiment, were used. They were operated in a David Kopf stereotaxic instrument under halothane anaesthesia. 6-OHDA (8 μg/4 μl), dissolved in 0.9% saline containing ascorbic acid (0.2 mg/ml), was injected bilaterally close to the nigro-striatal dopamine pathway at the level of lateral hypothalamus, 15 min after the rats had received protriptyline (10 mg/kg, IP), a noradrenaline uptake blocking agent. This type of procedure has been reported to cause a relatively specific degeneration of dopamine containing neurons [10]. The controls received the vehicle only. The coordinates, according to the stereotaxic atlas of König and Klippel [17] were A+3.4, L ± 1.3, V −3.2.

Ethanol intoxication was measured in these rats with a tilting-plane test [3], conducted blind 2 months after the operation. In this test the animal is placed on a wire-cloth covered plane, which is tilted by a motor at a constant speed from horizontal to vertical in 5 sec. The sliding angle of the rat is recorded. The rats were given a pre-ethanol test and then injected with 2 g/kg of ethanol IP as 12% w/v ethanol in 0.9% saline. Subsequent tilting-plane tests were performed 6 times at 20 min intervals after the injection. Each animal's intoxication on these tests was expressed as the percentage of its pre-ethanol performance shown after ethanol, using the sine of the sliding angle as the measure of performance. One-hundred-sixty min after the ethanol injection a blood sample of 0.05 ml was taken from the tip of the tail of each rat for gas chromatographic determination of the blood ethanol concentration [9].

RESULTS

All the 6-OHDA-treated rats developed severe akinesia, aphagia and adipsia, and had to be supported by tube feeding, which is an indication of substantial degeneration of the dopamine neurons [22]. A catalepsy test was performed two months after the operation by placing the rat's front paws on a 65 mm high block. The 6-OHDA-treated animals stayed in this position for 122 ± 24 sec (mean ± SEM); the controls stayed only 5 ± 2 sec ($t=3.810$; $df=24$; $p<0.001$).

The effect of the lesions on ethanol intoxication is shown in Fig. 1. The ethanol-induced impairment of performance on the tilting plane in the 6-OHDA-treated group was significantly less than in the controls throughout the experiment. There had been no difference in the performance of the 6-OHDA-group (59 ± 1 degrees; mean ± SEM) and the control group (58 ± 2 degrees) in the tilting plane test before ethanol treatment. The blood ethanol concentration of the

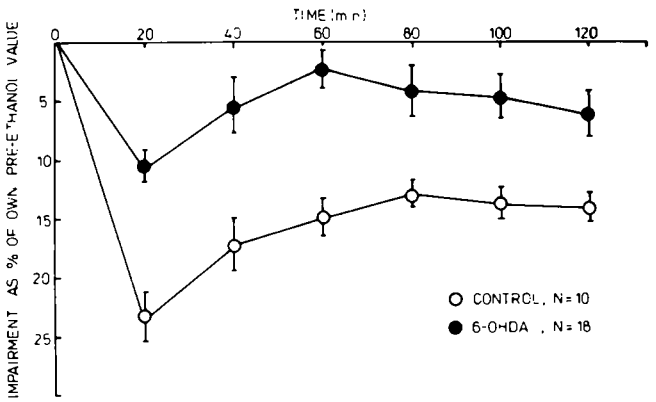

FIG. 1. Effect of a 6-hydroxydopamine (6-OHDA)-induced lesion of ascending dopamine pathways on ethanol intoxication in the rat. Each point represents the mean impairment of performance as the percent of the value (=sine of the sliding angle) before ethanol in the tilting plane test ± SEM in 6-OHDA-treated and control rats after a dose of 2 g/kg IP of ethanol. A profile analysis [19] revealed a highly significant difference in the level of performance between the groups ($t=5.513$; $df=26$; $p<0.001$).

6-OHDA-injected (38.5 ± 2.6 mM; mean ± SEM) was not significantly different from that of the controls (34.6 ± 0.9 mM).

DISCUSSION

The present finding that destruction of ascending dopamine pathways markedly inhibits ethanol induced intoxication in rats was not caused by a difference in the blood ethanol concentrations which were relatively similar in both groups in the end of the experiment. In fact the lesioned animals had slightly higher levels which should have made them more intoxicated rather than less. The differences also could not have been caused by only motor impairment, eg. akinesia, after 6-OHDA-treatment, because no difference was found between the groups in the performance before ethanol.

A relationship between ethanol and the dopamine systems has been previously suspected. An acute administration of ethanol has been shown to activate central dopamine neurons. Ethanol has been found to increase the formation of ^{3}H-dopamine from ^{3}H-tyrosine [5], which suggests increased dopamine synthesis, and to enhance significantly brain concentrations of the predominant metabolites of dopamine, homovanillic acid and 3,4-dihydroxyphenylacetic acid [15], which suggests increased dopamine metabolism. However, the dependence of any of the acute effects of ethanol upon dopamine has not been shown previously. It has been demonstrated that the ethanol-induced locomotor stimulation is prevented in mice and rats by α-methyl-p-tyrosine [4] and is partially restored by l-dopa in mice [7], and that α-methyl-p-tyrosine prolongs ethanol-induced sleeping time in mice [8]. However, the treatments used in these studies affect both brain dopamine and noradrenaline neurons, whereas the 6-OHDA treatment after protriptyline, used in the present study, is assumed to influence selectively the brain dopamine neurons [10].

There are 2 ways of interpreting the antagonistic effect of the lesion on ethanol intoxication. It can be speculated that dopaminergic neuronal systems are involved in the expression of the effects of an acute dose of ethanol and that the lesion, therefore, reduced expression of intoxication. On the other hand, it is possible that surviving neurons are able to compensate for the 6-OHDA-induced losses. Previous studies have provided evidence that there is compensatory enhancement in the dopamine turnover of the remaining dopamine neurons and increased sensitivity of the denervated post-synaptic receptors after 6-OHDA-induced dopamine lesion. Perhaps as a consequence the postsynaptic cells in the striatum show a several fold increase in mean unit activity after such a lesion [21,23]. Thus in terms of data suggesting an activating effect of acute ethanol on central dopamine systems [5,15], it is possible that the compensatory postsynaptic receptor supersensitivity and the alterations of dopamine turnover in the surviving dopamine neurons after subtotal 6-OHDA-induced loss of the dopamine neurons, in combination with the activating effect of ethanol on these neurons, could provide a basis for hyperfunction of the central dopamine systems. This in turn could inhibit the intoxicating effects of ethanol. This view is supported by findings that amphetamine, a drug which increases central dopaminergic tone [14], antagonized the ethanol-induced impairment in the behaviour of rats on the tilting plane [24]. In line with this interpretation are also the findings that in rats belonging to an alcohol-preferring strain the intoxicating effect of ethanol was smaller [18] and the brain dopamine content higher [1] than in rats avoiding alcohol.

It is, however, not necessary that the central dopamine neurons have the primary role in the expression of the action of acute ethanol in this experiment. For instance GABA-neurons may also be involved, because the dopamine- and GABA-systems in the central nervous system are known to interact [2,11] and because it has been shown that a GABA-activating drug, aminooxyacetic acid, potentiates, and a GABA-antagonist, bicuculline, diminishes ethanol intoxication in rats [12], and that ethanol-induced locomotor stimulation is suppressed by GABA-like drugs [6].

In conclusion, the present results show that selective degeneration of ascending dopamine neurons with 6-OHDA decreases ethanol-induced behavioural impairment. This could be caused either by a reduction of impulse flow in dopamine neurons or by hyperfunction of the remaining cells after the 6-OHDA-induced lesion.

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