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ANXIOLYTIC ACTIVITY OF SOME β-CARBOLINES IN RATS

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β-Carbolines are condensation products of indoles (in particular, serotonin) with aldehydes, they are found in plant and animal tissues [5], and they are of great importance for the regulation of certain processes of higher nervous activity, including the formation of alcohol motivation [3, 4]. The role of endogenous ligand of benzodiazepine receptors (BD-receptors) has been ascribed to certain natural β-carbolines [10, 11]. Meanwhile selective high-affinity binding of certain synthetic analogs of β-carbolines with BD-receptors has been described in certain regions of the brain, but the relationship between ligand and anxiolytic properties has not yet been adequately studied. The aim of the present investigation was accordingly to study this problem.

EXPERIMENTAL METHOD

Anxiolytic activity was studied by a method which is a variant of the conflicting situation: the appearance of motivated fighting between rats of a pair for safe territory (a bench above an electrode floor) during electronociceptive stimulation of the limbs. If both rats of the pair jump together onto the safe bench in the course of 1 min and remain on it for 10 sec, this is assessed as the manifestation of an anxiolytic effect [2]. Antineurotic action was studied as the ability of the drugs to influence an avoidance reaction formed in one rat during nociceptive stimulation of the other [1]. During investigation of the antineurotic effect of the compounds their doses were 5-8 times higher than ED_{50} values characteristic of the anxiolytic effect of the drugs [4].

Experiments were carried out on noninbred male albino rats weighing 180-200 g (antineurotic effect, effect of bicuculline) or weighing 480-500 g (anxiolytic action on a model of

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TABLE 1. Anxiolytic and Antineurotic Action of β-Carbolines

Compound	Anxiolytic effect	Antineurotic effect	Weakening of anxiolytic effect by bicuculline, %
	IC_{50}		
1-Me-6-MeO-THBC (1-methyl-6-methoxy-tetrahydro-β-carboline)	0,47 (0,36—0,62)	0 (1,0—65,0)*	no effect
1-Me-6-MeO-DBC (1-methyl-6-methoxy-dihydro-β-carboline)	0,015 (0,01—0,21)	0 (0,1—25)*	no effect
BC-3-CEE (β-carboline-3-carboxy-ethyl ester)	0,22 (0,17—0,28)	2,12 (1,51—2,96)	100
Diazepam	0,16 (0,11—0,24)	1,0 (0,76—1,3)	100

Legend. 0) Absence of effect. Confidence intervals of means at P = 0.05 shown in parentheses; dose ranges studied shown in parentheses with asterisk.

experimental alcoholism in rats). All the compounds tested were injected intraperitoneally in the form of an emulsion with Tween-80 30 min before the experiment began. Bicuculline (from Serva, West Germany), a specific blocker of GABA-receptors, was injected in a dose of 0.8 mg/kg (in this dose a 100% reduction of the effect of diazepam was observed). The experimental results were analyzed in an alternative form and by using the methods of Student, Litchfield, and Wilcoxon, with calculation of 50% effective doses.

EXPERIMENTAL RESULTS

A combination of anxiolytic and antineurotic activity with high sensitivity to the action of GABA antagonists, including to bicuculline, is very characteristic for diazepam and other tranquilizers of this series. The spectrum of pharmacological properties of BD-tranquilizers is due to their selective action on the GABA-BD complex. It follows from Table 1 that similar properties were possessed by BC-3-CEE, a compound with high affinity for BD receptors, comparable with diazepam (IC_{50} was 0.000007 and 0.000004 μM respectively) [5]. Meanwhile representatives of the unsaturated di- and tetrahydrocarbolines, which do not possess marked affinity for BD-receptors (IC_{50} is 0.21 and 0.15 mM respectively) [5], demonstrate a selective anxiolytic effect, with no antineurotic properties and without sensitivity to the action of bicuculline. At present at least two subtypes of BD-receptors have been identified: I) realizing the anticonvulsant action of benzodiazepines (main location the cerebellum), II) responsible for the anxiolytic effect (main location the cortex and hippocampus) [9]. Most β-carbolines, while exhibiting high affinity for type II BD-receptors, at the same time differ from BD-ligands in their sensitivity to GABA modulation and in other features discovered in radioreceptor investigations [6, 7]. This suggests the existence of specific β-carboline receptors, possibly coupled functionally and structurally with the GABA-BD complex. The results of the present investigation can be regarded as indirect confirmation of the existence of carboline receptors, by means of which their anxiolytic action is perhaps realized.

The marked change in function of the GABA-BD complex during the formation of experimental alcoholism and, in particular, in a state of abstinence [8], has enabled a model of this disease to be used to study the effectiveness of action of drugs inducing an anxiolytic effect, and differing in their degree of affinity for BD-receptors.

Investigations (Fig. 1) showed that the formation of experimental alcoholism involves a sharp fall in anxiolytic activity of diazepam and of BC-3-CEE, a highly effective liquid of BD-receptors.

Sensitivity to compounds with an anxiolytic action in rats with established dependence on ethanol (stage III) was thus reduced, for diazepam by 10.5 times compared with the con-

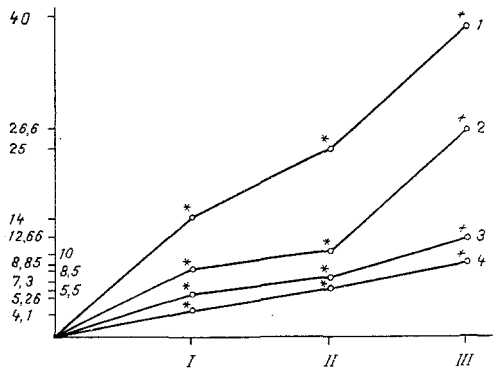

Fig. 1. Changes in sensitivity to anxiolytic action of compounds in rats with experimental alcoholism. Abscissa, stage of experimental alcoholism; ordinate A_1/A_0 (A_1 denotes ED_{50} of anxiolytic effect of compounds in rats at different stages of experimental alcoholism, A_0 — ED_{50} of anxiolytic effect of compounds in intact rats). 1) BC-3-CEE, 2) diazepam, 3) 1-Me-6-MeO-THBC, 4) 1-Me-6-MeO-DBC. I) Stage I, II) stage III, III) abstinence syndrome. *$P < 0.05$.

trol, and by derivatives of β-carbolines with low affinity for BD receptors by 5-7 times, and with high affinity by 25 times. The same rule, but more clearly emphasized, was found also in the period of abstinence. In this case the compounds studied were in the following order of effectiveness: β-carbolines (low affinity) > diazepam β-carbolines (high affinity).

The β-carbolines with low affinity for BD receptors gave a more stable effect in both stages I and III of experimental alcoholism and during the formation of an abstinence syndrome. Subpopulations of binding proteins, possibly β-carboline receptors, were evidently more resistant to the action of ethanol than the GABA—BD complex, which accounts for the great anxiolytic effectiveness of β-carbolines with low affinity for BD-receptors in experimental alcoholism. This investigation confirmed experimentally the reduction of effectiveness of tranquilizers exerting their action through the GABA—BD complex in alcoholism, and it suggests a purposive pharmacological search for agonists of "anxiolytic" systems coupled with BD-receptors or independent, among which may probably be included β-carboline systems.

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