PHYSIOLOGY

Peripheral Administration of Loperamide and Methylnaloxone Decreases the Degree of Anxiety in Rats

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We studied the effect of μ-opioid receptor ligands on anxious and depressive behavior of rats. Intragastric administration of loperamide and methylnaloxone reduced animal anxiety evaluated by an increase in the number of entries into and time spent in open arms of the elevated plus-maze. μ-Opioid receptor agonist loperamide had the most pronounced anxiolytic effect. Analysis of animal behavior in the forced swimming test showed that administration of μ-opioid receptor antagonist methylnaloxone reduced the latency of the first submersion, increased the total time of submersion episodes, and shortened the time of active swimming, which attested to depressive properties of this agent. Loperamide had little effect on behavior of rats in the forced swimming test. Thus, μ-opioid receptor agonist loperamide has the anti-anxiety properties and produced no sedative effect. Therefore, this agent holds much promise as an anxiolytic drug.

Key Words: *central and peripheral μ-opioid receptors; methylnaloxone; loperamide; anxiolytic effect; elevated plus-maze*

High incidence of anxious disorders and low efficacy of medicinal products necessitate the search for new neuronal targets for original anxiolytic drugs [1,11-13]. Apart from a variety of neurotransmitter systems, the endogenous opioid system of the brain plays an important role in the mechanisms of emotional behavior [8,10]. The structure of opioid receptors and endogenous opioid peptides is practically similar in CNS and peripheral tissues. At the same time, the central and peripheral functions of the endogenous opioid system are different due to impermeability of the blood-brain barrier (BBB) for most opioid peptides [7]. Therefore, the central and peripheral functions of the endogenous opioid system are studied separately [9]. Our recent experiments showed that μ-opioid receptor ligands not crossing BBB have different effects on the density of specific receptors in the cerebral cortex of intact rats [3]. These data and results of studying the central effect of peripheral treatment with opioid receptor ligands suggest that activation of the peripheral compartment can inhibit the central compartment of the opioid system, and vice versa [5].

Here we studied the effect of μ-opioid receptor ligands on depressive and anxious behavior of rats.

MATERIALS AND METHODS

Experiments were performed on 24 male Wistar rats weighing 180-230 g. The animals were housed in

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cages (8 rats per cage) and had free access to water and standard combined food. The experiment was conducted in accordance with the Order No. 267 of the Russian Ministry of Health (19.06.2003) and "Rules of Studies on Experimental Animals" (P. K. Anokhin Institute of Normal Physiology; protocol No. 1, 03.09.2005). The rats were divided into 3 groups (8 animals per group). Group 1-3 animals received intragastrically (through a tube) loperamide (5 mg/kg; Sigma), naloxone methiodide (5 mg/kg; Sigma), and distilled water (equivalent volume), respectively. Anxiolytic activity of compounds was evaluated using the elevated plus-maze (EPM) test as described previously [14]. Each rat was placed in the center of EPM 30 min after administration of study compounds. The behavior of animals was studied by the standard method for 5 min. Depressiveness was estimated in the Porsolt forced swimming test. The rat was placed in a swimming pool (glass cylinder, 54 cm in height, 46 cm in diameter, water temperature 25-26°C) 30 min after treatment. The following parameters of behavior were recorded over 5 min: duration of immobility; number of immobility periods; time of active swimming; *etc*.

The results were analyzed by analysis of variance (ANOVA).

RESULTS

Administration of loperamide and methylnaloxone was followed by a decrease in the anxiety of animals. The most significant changes were observed after treatment with μ-opioid receptor agonist loperamide. Similarly to methylnaloxone, loperamide significantly increased the number of entries into and time spent in the open arms. These changes reflect a strong anxiolytic effect of the test compounds (Fig. 1). Other behavioral parameters in treated rats did not differ from those in control animals.

Analysis of animal behavior in the forced swimming test showed that administration of μ-opioid receptor antagonist methylnaloxone decreased the latency of the first submersion, increased the total time of submersion episodes, and shortened the time of active swimming. These changes were not observed under the influence of a μ-opioid receptor agonist loperamide (Fig. 2).

Loperamide produced a strong anxiolytic effect; the depressive was insignificant. Peripheral administration of methylnaloxone was mainly accompanied by a strong depressive effect and slight anxiolytic action.

Our recent experiments showed that peripheral administration of methylnaloxone increased the release of β-endorphin from nerve endings in the rat brain. These changes are accompanied by an increase in the number of μ-opioid receptors [3,4]. The antianxiety effect of methylnaloxone is probably related to these changes. Loperamide slightly inhibits the release of β-endorphin and decreases the number of μ-opioid receptors in the cerebral cortex. However, emotional stress is followed by massive release of β-endorphin (as distinct from the effect of methylnaloxone). The observed changes probably contribute to the antianxiety and antistress effect of loperamide.

Published data show that anxiolytic agents have sedative and depressive side effects [1,2,6,15]. The results of our study indicate that a μ-opioid receptor agonist loperamide produces a strong antianxiety effect and exhibits no sedative activity. We conclude that loperamide holds much promise as an anxiolytic drug with an original mechanism of action.

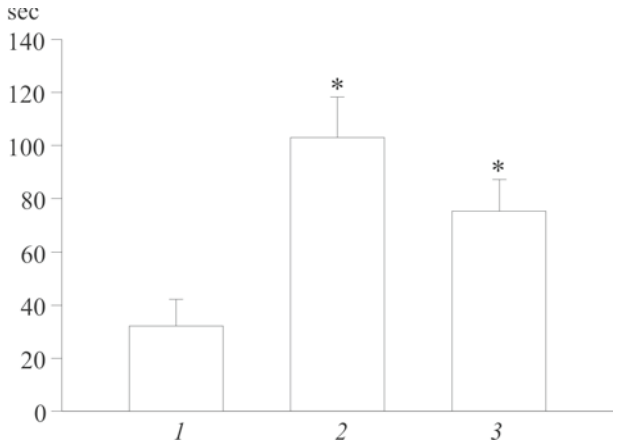

Fig 1. Time spent in the open arms of EPM after intragastric administration of distilled water (*1*), loperamide (*2*), and methylnaloxone (*3*). Here and in Fig. 2: *p<0.05 compared to distilled water.

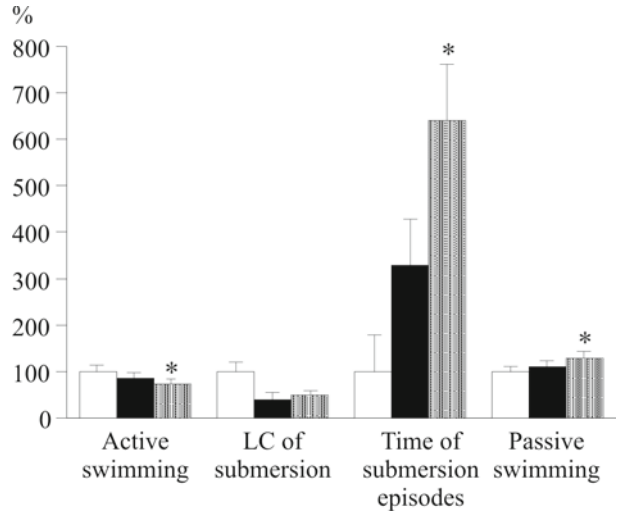

Fig. 2. Period of active swimming, latency (LC) of the first submersion, total time of submersion episodes, and total time of passive swimming in the forced swimming test after intragastric administration of distilled water (light bars), loperamide (dark bars), and methylnaloxone (shaded bars). Time parameters in the control group are taken as 100%.

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