PHYSIOLOGY

Hypothesis on Reciprocal Interactions between the Central and Peripheral Components of the Endogenous Opioid System

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A hypothesis on reciprocal interactions between the central and peripheral components of the endogenous opioid system was formulated on the basis of results of our experimental studies and published data. In order to verify this hypothesis, we studied the effects of peripheral administration of loperamide (μ-opioid receptor agonist) and methylnaloxone (opioid receptor antagonist) not penetrating through the blood-brain barrier on the pain sensitivity of rats, morphine-induced analgesia, and formation of morphine analgesia tolerance. Peripheral loperamide and methylnaloxone modulated the central mechanisms of perception of painful stimuli. This fact confirmed the hypothesis on the reciprocal interactions between the central and peripheral compartments of the endogenous opioid system. Methylnaloxone exhibits an antagonistic effect on peripheral μ-opioid receptors, which probably leads to activation of the central μ-opioid receptors and to the development of analgesia. Loperamide activates peripheral, but suppresses the central μ-opioid receptors, which leads to hyperalgesia. Methylnaloxone suppresses morphine-induced analgesia under conditions of morphine activation of the central antinociceptive mechanisms. Peripheral injection of μ-opioid agonist loperamide virtually did not modify the central compartments of the opioid system under conditions of morphine treatment. Methylnaloxone and loperamide partially prevented the development of morphine analgesia tolerance. Hence, the results confirm the hypothesis about the reciprocal interactions between the central and peripheral compartments of the endogenous opioid system. The relationships between the central and peripheral compartments of the opioid system can be more intricate when its function is modulated.

Key Words: *central and peripheral opioid receptors; methylnaloxone; loperamide; pain sensitivity; analgesia*

The endogenous opioid system was discovered and described in the 1970s. It is known that various compartments of the CNS and many peripheral organs and tissues have opioid receptors of three types: μ, δ, and κ. Endogenous opioid ligands of these receptors are endorphins, enkephalins, and dynorphins. The structure of receptors and opioid peptides is the same in CNS and at the periphery, but the central and peripheral functions of the endogenous opioid system are different, because the majority of opioid peptides cannot penetrate

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through the blood-brain barrier (BBB). That is why the central and peripheral functions are studied separately. The effects of exogenous opioid receptor agonists and antagonists penetrating through BBB were described not once. The majority of changes caused by them are due to their effects on the CNS and the periphery. Few studies of the central effects of peripheral administration of opioid peptides or other opioid receptor agonists or antagonists, not penetrating through the BBB gave paradoxical results. Subcutaneous injection of conjugated β-endorphin stimulated alimentary behavior [2] and suppressed the sensitivity to the analgesic effect of morphine [1]. Epileptiform activity in the brain cortex in rats receiving a peripheral injection of β-endorphin analog was suppressed [7]. Intraperitoneal injection of β-endorphin analog lacking opioid activity stimulated morphine analgesia in mice [12] and rats [11]. Peripheral injection of a fragment of β-endorphin (l-27) suppressed (similarly to naloxone) stress-induced analgesia [13]. Moreover, injection of ethanol to C57Bl/6 mice increased plasma level of β-endorphin and decrease it in the hypothalamus [8].

Experiments performed by us over many years yielded results which are difficult to interpret from the common viewpoint. It was found, for example, that peripheral injections of endopeptidase inhibitors antipain and leupeptin modified pain sensitivity and morphine analgesia, suppressed animal sensitivity to morphine positive reinforcement, and suppressed the abstinence syndrome in morphine-dependent rats [10]. Antipain and leupeptin are small peptide molecules, however it was reported that they could not penetrate through the BBB. The abstinence syndrome was also suppressed after intraperitoneal injection of peptidases inhibitor aprotinin, a high-molecular-weight polypeptide *a priori* not penetrating through BBB [3]. It was found that blood level of β-endorphin was elevated in rats with opium abstinence, while aprotinin normalized its level [6]. However, we failed to explain at that time the paradoxical fact that the decrease in the level of β-endorphin could inhibit the abstinence syndrome. Later we studied the effect of active immunization of animals with monoclonal antibodies to morphine. We found that the rats produced anti-idiotypical antibodies to morphine binding to the peripheral (but not central) opioid receptors, because the Ig could not penetrate into the CNS. Immunized rats differed from normal ones by increased sensitivity to the analgesic and decreased sensitivity to the reinforcing effect of morphine [4]. Based on these facts (and published data), we put forward a hypothesis on reciprocal interactions between the central and peripheral components of the endogenous opioid system; in other words, we hypothesized that inhibition of peripheral activity leads to activation of the central compartment and vice versa. In order to verify this hypothesis, we studied the effect of peripheral administration of μ- and δ-opioid receptor antagonists on the severity of the abstinence syndrome in morphine-dependent rats. Naloxone easily penetrating through BBB sharply stimulated the abstinence syndrome, while methylnaloxone not penetrating into the CNS inhibited it [5].

We studied the effects of peripheral methylnaloxone and loperamide on pain sensitivity in rats, morphine-induced analgesia, and formation of morphine analgesia tolerance.

MATERIALS AND METHODS

Experiments were carried out on 60 male Wistar rats (180-200 g). The rats were kept 8-10 per cage with free access to water and food.

Pain sensitivity was evaluated by the tail flick test: the tail (5 cm) was placed into hot water (56°C) and the latency of tail withdrawal was recorded. The initial tail flick latency was measured in 30 rats. Ten animals of each group were then intraperitoneally injected with isotonic NaCl, naloxone metiodide (10 mg/kg; Sigma), or loperamide (10 mg/kg; Sigma), respectively. The tail flick latency was evaluated 10, 30, 60, 120, and 180 min after injection. Morphine analgesia was evaluated in 30 rats in the same test after preinjection (intraperitoneal) of morphine hydrochloride (5 mg/kg). During the next 2 days, the animals of this series received additional single injections of morphine. On day 3, the tail flick test was repeated in order to evaluate morphine analgesia tolerance.

The data were processed by the ANOVA.

RESULTS

Intraperitoneal methylnaloxone exhibited a significant analgesic effect, while loperamide reduced the tail flick latency (Fig. 1).

Injection of morphine markedly prolonged tail flick latency. Maximum analgesia (up to 500%) was observed 60 min after morphine injection, after which the tail flick latency returned to the initial level. Slight, but significant hyperalgesia was recorded 120 min after morphine injection. Intraperitoneal loperamide virtually did not modify the analgesic effect of morphine. By contrast, intraperitoneal methylnaloxone significantly inhibited morphine-induced analgesia (Fig. 2). It is noteworthy that both methylnaloxone and loperamide exhibited significant analgesic effects during the second hour after

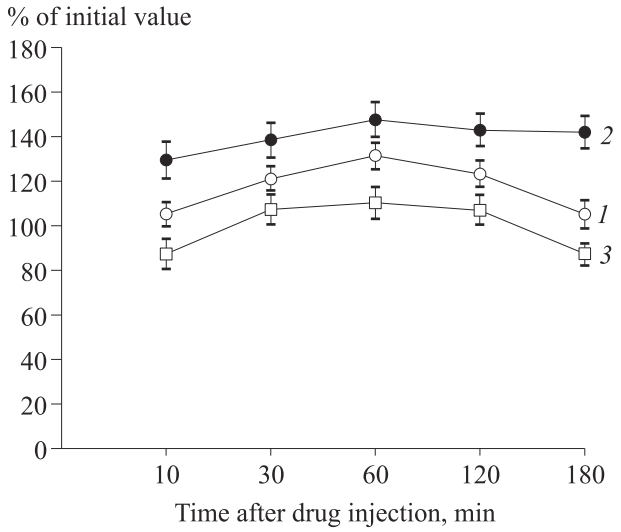

Fig. 1. Pain sensitivity in rats injected with isotonic NaCl (*1*), methylnaloxone (*2*), and loperamide (*3*). Here and in Fig. 2: ordinate: tail flick latency.

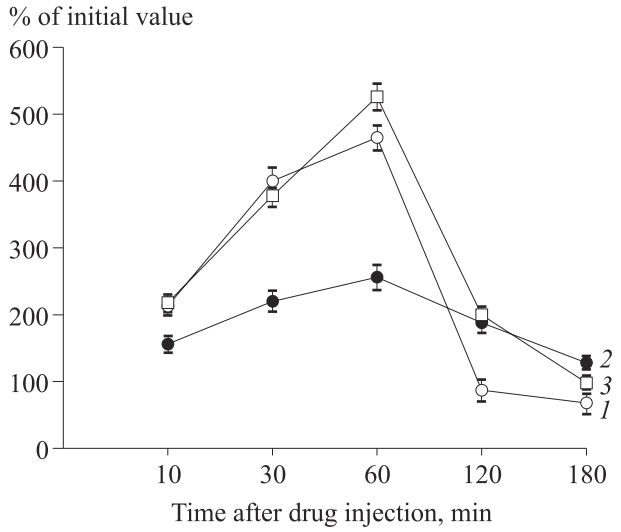

Fig. 2. Pain sensitivity in rats injected with morphine in combination with isotonic NaCl (*1*), methylnaloxone (*2*), and loperamide (*3*).

their injections, when the analgesic effect of morphine was over.

It is known that repeated injections of morphine induced the development of tolerance of its analgesic effect. In our experiments, the analgesic effect of morphine decreased by 45% on day 3 of injections in comparison with day 1. Peripheral injection of loperamide resulted in just a 17% reduction of morphine analgesic effect on day 3. After injection of methylnaloxone, the analgesic effect of morphine was weakened by 30% (Fig. 3).

Hence, peripheral loperamide and methylnaloxone modulated the central mechanisms of perception of pain stimuli. This fact confirms the hypothesis about reciprocal interactions between the central and peripheral compartments of the endogenous opioid system. Methylnaloxone really exhibited the antagonistic effect on the peripheral μ-opioid receptors, which seemed to lead to activation of the central μ-opioid receptors and to the development of the analgesic effect. By contrast, loperamide, activating the peripheral μ-opioid receptors, suppressed the central ones, which caused hyperalgesia.

It seems that the effects of peripheral compartment of the opioid system on its central compartments depend on functional state of the latter ones. We previously discussed the possibility of activation of the central opioid system, suppressed during the morphine abstinence syndrome, by injection of peripheral antagonist (methylnaloxone) [5]. The results of the present study indicate that under conditions of potent activation of the central antinociceptive mechanisms by morphine, methylnaloxone suppresses this activation. Peripheral μ-opioid agonist loperamide virtually did not modify the central compartments of the opioid system under conditions of morphine treatment. It is known that the formation of morphine tolerance and, specifically, to its analgesic effect, can be associated with the mechanisms of adaptation of the opioid receptors located in the antinociceptive compartments of the brain [9]. It can not be excluded that suppression of the peripheral opioid system with methylnaloxone led to activation of adapted central opioid receptors, this stimulating the analgesic effect of morphine in rats with morphine tolerance. Presumably, repeated treatment with loperamide in combination with morphine led to pronounced adaptation processes in the peripheral opioid receptors, which caused activation of the central receptors and suppressed tolerance formation.

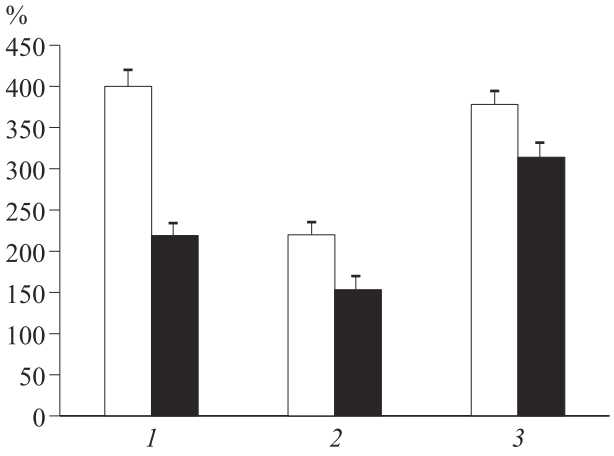

Fig. 3. Analgesia (ratio of latency 30 min after morphine injection to initial tail flick latency, in %) after injection of isotonic NaCl (*1*), methylnaloxone (*2*), and loperamide (*3*) on days 1 (light bars) and 3 (dark bars) of the experiment.

Hence, the hypothesis on the reciprocal interactions between the central and peripheral compartments of the endogenous opioid system was confirmed. Modification of the opioid system function can lead to more intricate shifts in the proportion of its central and peripheral compartments.

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