ROLE OF CATECHOLAMINE NEURONS OF LATERAL RETICULAR NUCLEI IN PAIN SENSITIVITY CONTROL DURING REFLEX STIMULATION

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Electrophysiological investigations [2, 3, 6] have shown that an important role in the depression of pain sensitivity by the action of opiates or electrical stimulation of various brain structures is played by descending spinal inhibitory systems projecting from the brain stem. Besides serotonin systems, catecholaminergic mechanisms also are involved in the mechanisms of depression of pain sensitivity under the influence of opiates or electrical stimulation. Among these brain structures may be included the lateral reticular nuclei (A-1), whose catecholaminergic neurons give projections to the posterior horns of the spinal cord [5]. It has in fact been shown that the analgesic effect of morphine is considerably reduced in rats by blocking catecholamine neurons in nuclei A-1 [8], evidence of the role of these brain structures in the regulation of pain sensitivity. However, the role of nuclei A-1 in mechanisms of analgesia in procedures such as acupuncture and stimulation of organs of the pelvis minor (SOPM), which induce a marked analgesic effect, has not yet been explained.

It was accordingly decided to study the effect of specific pharmacological blocking of catecholamine-containing neurons in nuclei A-1 of the rat brain on the time course of pain sensitivity during auricular electroacupuncture and SOPM.

EXPERIMENTAL METHOD

Altogether 30 albino rats (13 males and 17 females) weighing 200-250 g were used. The animals were anesthetized with chloral hydrate (4.5 ml/kg of an 8% solution, intraperitoneally) and fixed in a **stereotaxic** apparatus. Through a cannula connected to a microsyringe by means of a polypropylene tube, the position of which corresponded to coordinates AP 6.0, L ±2.0, VD −3.2 mm [7], a solution of 6-hydroxydopamine (6-OHDA, 9 μg/2 μl/min, from Sigma, USA) was injected into the experimental rats, in which it caused specific damage to catecholamine neurons. Sterile physiological saline was injected into the control animals. All solutions contained ascorbic acid (0.2 mg/ml). Experiments were carried out 10-12 days after the operation. To induce analgesia, auricular electrical stimulation (AES) was applied in the "lung" region through clip electrodes, with a pulsed current of 0.6-1.0 mA, 4 Hz, 0.04 msec, for 5 min and SOPM was applied by the method in [4]. For this purpose the glass plunger of a tuberculin syringe was inserted into the vagina of the female rats under a pressure of 600-700 g for 5 min. As a result of this procedure the rats developed total immobility, accompanied by depression of pain sensitivity equivalent to the action of morphine in a dose of 2 mg/kg.

In the experiments with AES pain sensitivity was evaluated by measuring latent periods of the **paw-licking** response of rats placed on a hot plate at 55°C (hot plate test, HPT) and the tail-flick test (TFT) in response to the action of a focused beam of light from a 150-W projection lamp. In the experiments with SOPM only results of the TFT were used. Responses to pain in the HPT and TFT were measured 7-10 min before the beginning of the test and at definite time intervals thereafter.

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The brain was removed from five rats 24 h after the operation and fixed in 10% neutral formalin solution. Sections 60 μ thick were then cut on a freezing microtome and stained by the standard method with cresyl violet to verify that the cannula was in nucleus A-1. The remaining animals of the experimental and control groups were killed 24-36 h after the experiment. The brain and spinal cord were frozen at −30°C in dry ice and kept for 1.5-2 weeks in a refrigerator at −20°C. Concentrations of noradrenalin and serotonin in brain and spinal cord tissues were determined microfluorometrically [1].

EXPERIMENTAL RESULTS

The results of experiments to study the action of AES are given in Fig. 1. Sensitivity to pain, measured by the HPT and TFT before AES, was the same in rats of the experimental and control groups. The latent period of the HPT was 7.7 ± 1.1 sec in the experimental and 8.2 ± 1.4 sec in the control rats; those of the TFT were 2.3 ± 0.1 and 2.4 ± 0.3 sec, respectively. This fact is evidence that injury to catecholamine neuronal systems of nuclei A-1 does not affect the mechanisms controlling sensitivity of rats to pain at rest. Under the influence of AES a significant increase in the response to HPT was observed in the control rats and it lasted 10 min. By contrast, in the experimental animals the results of HPT began to be even lower after the AES than before it. Differences between the experimental and control series were significant.

Similar results were obtained when sensitivity to pain was evaluated by the TFT (Fig. 1). AES of zones of acupuncture points on the concha auriculae of the control rats was accompanied by significant lengthening of the latent period of the TFT, which continued for about 30 min. In the experimental group the values fell below the initial level. Comparison of their values in the two groups showed a significantly lower value in the experimental rats.

TABLE 1. Noradrenalin and Serotonin Concentrations (in % of control) in Rats after Injection of 6-OHDA into Lateral Reticular Nuclei

Procedure	Test object	Noradrenalin	Serotonin
AES	Spinal cord	55	96
	Brain "	105	—
SOPM	Spinal cord	61	94
	Brain "	106	—

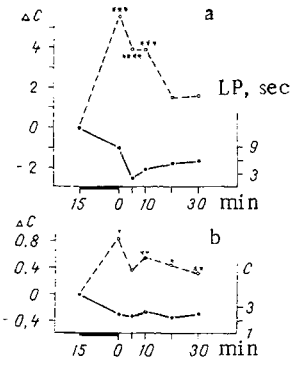

Fig. 1

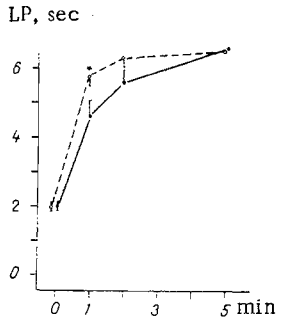

Fig. 2.

Fig. 1. Effect of blocking catecholamine systems of nuclei A-1 on latent periods of HPT (a) and TFT (b) and their time course after AES. Broken line — control, continuous line — experiment. *P < 0.05, **P < 0.03, ***P < 0.001, ****P < 0.002 compared with control. Time course of latent periods shown in relative units.

Fig. 2. Effect of blocking of catecholamine systems of nuclei A-1 by 6-OHDA on latent periods of TFT before and during visceral stimulation. Broken line — control, continuous line — experiment. *P = 0.05.

When SOPM was used, this procedure caused a significant decrease in sensitivity to pain in both the control and experimental series, as shown by the sharp increase in values of the TFT (Fig. 2). Comparison of results for the groups showed a significant difference ($P = 0.05$) only toward the end of the 1st minute, with a shorter TFT result in rats receiving 6-OHDA. At all other time intervals no difference was found in the latent periods of the different groups.

The histological control showed that the tip of the cannula was located in the region of the lateral reticular nuclei. Evidence that the descending catecholamine pathways of the spinal cord were blocked was given by the results of biochemical analyses (Table 1). In animals receiving 6-OHDA the noradrenalin concentration was significantly reduced only in the spinal cord, and its level in the brain was unchanged. The serotonin concentration was about equal in tissues studied from the experimental and control animals.

The following conclusion can thus be drawn from these results. First, blocking noradrenergic systems of nuclei A-1 does not affect the initial sensitivity of rats to pain, as shown by the fact that the initial values of latent periods of the HPT and TFT in the experiment and control did not differ. The reason may perhaps be that the "power" of the remaining noradrenergic neurons was sufficient to provide for mechanisms controlling sensitivity to pain. However, during functional loading of these mechanisms by the reflex procedures used in these experiments quantitative and qualitative differences in the activity of the analgesic systems were revealed. Second, reduction of the duration of responses to pain in the experimental rats compared with the control indicates that the catecholamine systems of nuclei A-1 and the descending systems of the spinal cord play an important role in the mechanisms controlling sensitivity to pain during AES and SOPM. Third, since a significant increase in the latent periods of the TFT was observed as a result of SOPM compared with their initial values, it can be postulated that besides noradrenergic systems, other neurochemical mechanisms, which may perhaps be connected with the functioning of different brain structures, also participate in the mechanism of activation of antinociception.

LITERATURE CITED

1. B. M. Kogan and N. V. Nechaev, Lab. Delo, No. 5, 301 (1979).
2. G. Belcher, R. W. Gyall, and R. Shaffner, Brain Res., 151, 307 (1978).
3. W. R. Crowley, J. F. Rodriquez-Sierra, and B. R. Komisaruk, Brain Res., 137, 67 (1977).
4. A. Dahlstrom and A. Fuxe, Acta Physiol. Scand., 64, Suppl. No. 247, 5 (1965).
5. J. G. Hall, A. W. Duggan, S. M. Johnson, et al., Neurosci. Lett., 25, 25 (1981).
6. L. S. Pellegrino, A. Pellegrino, and A. Cushman, A Stereotaxic Atlas of the Rat Brain, New York (1979).
7. A. Pert, J. Massari, Y. Tizabi, et al., in: Endogenous and Exogenous Opiate Agonists and Antagonists, E. L. Way, ed., Oxford (1980), pp. 151-154.