PHYSIOLOGY

Effect of Social Isolation on Behavioral Parameters and Sensitivity of Rats to Morphine and Caffeine

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Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 135, No. 1, pp. 11-13, January, 2003
Original article submitted October 16, 2002

Social isolation is a stress factor increasing animal anxiety and impairing food-reinforced instrumental learning. Social isolation modulates sensitivity to psychoactive substances: it potentiated the depressive and analgesic effects of morphine, but attenuated the activating and anxiogenic effects of caffeine. These behavioral changes and changes in sensitivity to psychoactive agents can be explained by a well-known phenomenon of activation of the endogenous opioid system during stress.

Key Words: *social isolation; learning; anxiety; analgesia; motor activity; morphine; caffeine*

Social isolation (SI) of an adult animal is a potent stress factor modulating activity of various physiological systems in the organism. In mice and rats SI led to the release of endogenous opioids and a decrease of opioid receptor sensitivity [12]. This, in turn, induced secretion of corticotropin and corticotropin releasing factor [2], which caused an increase of corticosterone level [3]. No doubt, these processes modify the behavior of animals, changing the level of their emotions [8,14], disturbing learning and memory [7], motivation [1], and reinforcement system [6,9].

The data on the effects of SI and other types of stress on sensitivity to psychoactive agents are contradictory. It was shown that stress increased the sensitivity to narcotics and stimulated narcotic consumption, for example morphine consumption [5,10], decreased [12,13] or potentiated [15] the analgesic effect of morphine, increased the sensitivity to morphine effect on motor activity [4], and decreased the sensitivity to its effect on body temperature [13]. Prenatal stress increases rat sensitivity to the activating effect of caffeine [11].

We investigated the effect of SI on emotional status of animals, their learning capacity, and sensitivity to psychoactive agents with activating (caffeine) and depressive (morphine) effects.

MATERIALS AND METHODS

Experiments were carried out on 96 male WAG/G rats (180-200 g). After the rats were received from the breeding center, half of them were immediately placed into individual 25×10×20 cm cages. Other animals were kept in groups (n=7-8) in 40×30×15 cm cages. Animals were kept under 12:12 h day-night regimen at 21°C with free access to standard fodder and water. Experimental procedures were carried out during the light phase at 11.00-16.00.

On day 28 the level of anxiety was evaluated in 16 rats (8 kept individually and 8 in groups) in the dark/light box test. An experimental box (35×16×21 cm) was divided into 2 equal compartments (illuminated and dark) with a hole for passage. The rats were placed into the light compartment and the latency of the first passage into the dark compartment and the

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0007-4888/03/1351-06$25.00

total time spent in the light compartment were measured for 5 min.

After 3 days the level of anxiety of the same rats was evaluated in Vogel conflict test. Before testing the rats were subjected to 72-h water deprivation. The animals were placed into experimental cage with access to water; each 10th touching of the drinking bowl was associated with an electric impulse (5 mA). The number of electric impulses received by the animals for 5 min was counted.

In other 16 rats, 8 of which were kept individually and 8 together for 28 days, learning capacity was studied in an instrumental food-getting paradigm. After 48-h food deprivation the rats were placed in an instrumental box (Lafayette Instruments Inc.), where the animal could get a 45-mg fodder granule (P. S. Noyes Company Inc.) after pressing a lever. The moment when the fodder was delivered into the feeder was followed by a 17-sec latent period (LP), when lever pressing was ineffective. LP was accompanied by switching off the light in the experimental box. The number of effective lever pressings was counted.

Behavior in the open field (OF) test and pain sensitivity before and after caffeine or morphine injection were studied in 64 rats. The animals were placed into a round OF 120 cm in diameter for 5 min. LP of motor activity and its intensity, duration of passive behavior, grooming reactions, number of excursions into the center of OF and rearings were counted. After 25 min LP in the tail-flick test (hot water, 56°C) was measured.

Ten min before testing 8 animals kept individually and 8 kept in group for 28 days received caffeine (50 mg/kg intraperitoneally), 8 animals received morphine hydrochloride (5 mg/kg intraperitoneally), and other animals received isotonic NaCl solution.

At the start of the experiment, 8 animals kept individually and 8 kept in group were deprived of water and offered 1% caffeine as the only source of fluid. After 28 days animal behavior was studied in a dark/light box 10 min after injection of isotonic NaCl.

The results were processed using Student's *t* test for non-paired cases.

RESULTS

SI increased the level of anxiety in rats. The rats kept in group spent 61.1±9.9 sec in the light compartment, while the rats exposed to SI spent there only 28.5±13.2 sec.

Anxiety of WAG/G rats evaluated by Vogel test was also higher in animals kept individually. The number of approaches to the drinking bowl was 69.3±11.2 in rats kept in a group and 41.1±7.9 after 31-day SI.

Social isolation led to significant inhibition of instrumental food-getting behavior: 12.8±5.5 *vs.* 32.9±9.0 effective lever pressings in rats kept in group and individually, respectively.

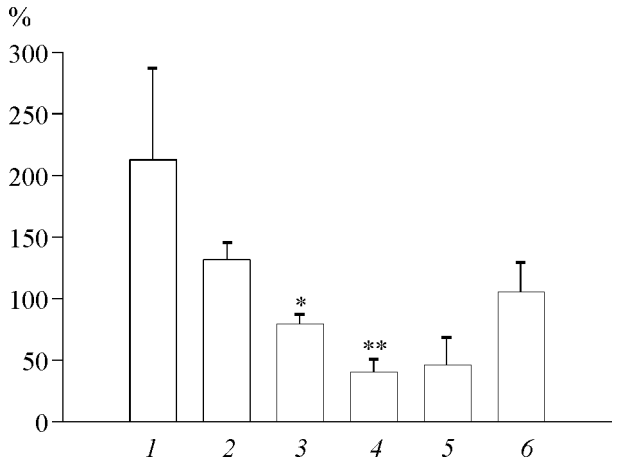

Fig. 1. Effect of social isolation on behavioral parameters in the open field test (*vs.* rats kept in group). *1*) latent period of motor activity; *2*) intensity of motor activity; *3*) duration of passive behavior; *4*) number of grooming reactions; *5*) number of excursions into the center of the field; *6*) rearings. *$p<0.05$, **$p<0.01$ *vs*. animals kept in group.

SI did not considerably change animal behavior in OF, but in rats kept individually motor activity was somewhat higher, the number of entries into the central zone and the time of grooming reactions decreased (Fig. 1).

Caffeine (50 mg/kg) produced an activating effect, while morphine (5 mg/kg) produced a slight depressive and analgesic effects. Motor activity in OF increased significantly after caffeine and decreased after morphine injection (Fig. 2). SI suppressed the activating effect of caffeine by 12.5% (the effects became negligible) and potentiated the depressive effect of morphine by 18.6% (Fig. 2, *b*).

SI did not change pain sensitivity: LP in the tail-flick test was 2.7±0.3 sec in animals kept in the group

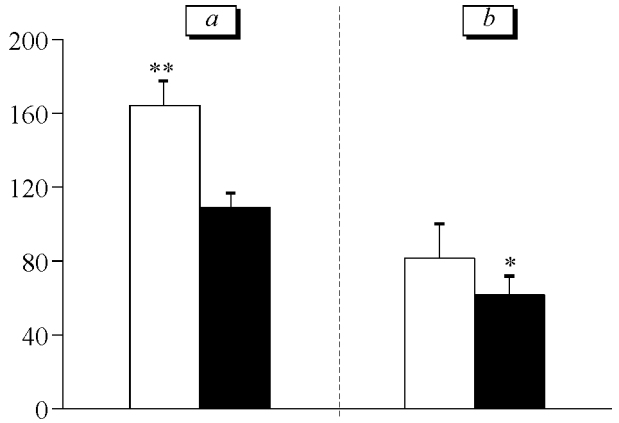

Fig. 2. Motor activity of rats kept in group (*a*) and individually (*b*) in the open field test after injection of caffeine (light bars) and morphine (dark bars). Ordinate: % of control (NaCl injection). *$p<0.05$, **$p<0.01$ *vs.* control.

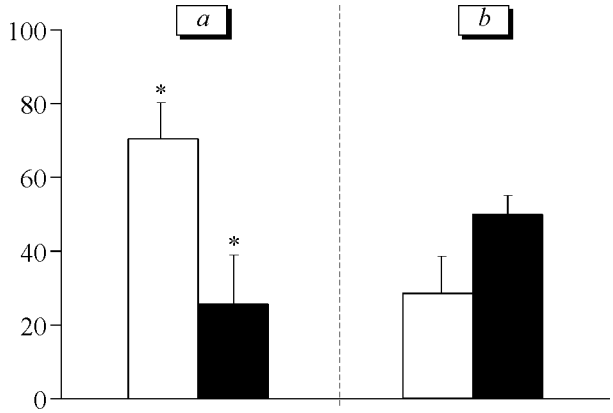

Fig. 3. Suppression of anxiogenic effect of caffeine in rats kept in a group (*a*) and individually (*b*). Light bars: water consumption; dark bars: caffeine consumption. Ordinate: time spent in the light compartment, sec. *$p<0.05$ compared to rats drinking water.

and 3.0±0.2 sec in those kept individually. Caffeine did not modulate the pain sensitivity in rats. The analgesic effect of morphine was 27.5% higher in rats kept individually.

Rats chronically drinking caffeine developed a trend to an increase of anxiety level. These animals spent less time in the light compartment. SI which increased anxiety, prevented the development of this process in animals chronically exposed to caffeine (Fig. 3).

Hence, SI is a stress exposure increasing animal anxiety and impairing learning. It modulates animal sensitivity to psychoactive agents: potentiates the effects of morphine and attenuating those of caffeine. These changes in behavior and sensitivity to psychoactive agents can be explained by the well-known phenomenon of activation of the endogenous opioid systems in stress.

The study was supported by the Russian Foundation for Basic Research (grant No. 01-04-48136).

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