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Different Effects of Short- and Long-Term Treatment With Imipramine on the Apomorphine- and Food-Induced Place Preference Conditioning in Rats

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PAPP, M. *Different effects of short- and long-term treatment with imipramine on the apomorphine- and food-induced place preference conditioning in rats.* PHARMACOL BIOCHEM BEHAV **30**(4) 889–893, 1988.—The effect of imipramine (IMI) on the rewarding properties of pharmacological and natural reinforcers was studied with a place preference paradigm. The pairing of distinctive environmental stimuli with either injection of different doses of apomorphine (APO) or presentation of food to hungry rats resulted in a conditioned preference for those stimuli. The development of APO- and food-induced place preference was prevented by short-term administration of IMI. In contrast, long-term pretreatment with IMI significantly potentiated the APO- and food-induced conditioned effect. In separate experiments aversive properties of IMI by itself were also found, as the conditioned avoidance response to the environmental stimuli paired with administration of single doses of IMI was demonstrated. The results indicate an enhancing effect of the long-term IMI administration on the dopamine-mediated reward functions and the rewarding value of natural reinforcers.

Imipramine Dopamine Reward Apomorphine and food reinforcement Rat

A growing number of studies have recently focused on a possible role of the central dopamine (DA) system in the pathogenesis of depression and in the mechanism of action of antidepressant drugs (AD). In animal studies, prolonged administration of both typical and atypical AD, repeated application of electroconvulsive shock and REM sleep deprivation resulted in enhancement of a variety of behavioural responses to DA agonists given either systemically or directly into brain structures [3, 15, 25, 26]. The postulated mechanism of this effect is the hypersensitivity of postsynaptic DA receptors in the mesolimbic system, developed as a result of antidepressant therapy [14,30]. There are also some evidences for the hyposensitivity of presynaptic DA receptors in this system in animals chronically treated with AD [23,24], but other authors do not confirm these findings [4,26]. Since the dysfunction of DA system—at least in some kinds of depression— has been supported by clinical studies (see [29]), the interest in this system, as regards its involvement in the therapeutic effects of AD, seems to be well-founded.

A number of data point to the involvement of central DA-containing neurons in the control of animal reward functions. The lesion of these neurons by 6-hydroxydopamine, as well as the blockade of DA receptors by neuroleptics, attenuate the rewarding properties of both natural and artificial stimuli. On the other hand, under various schedules of reinforcement, the animal response is enhanced by facilitation of the neurotransmission at DA synapses [5, 20, 21, 31]. These results are also confirmed by human studies showing the enhanced positive aspects of mood as a result of DA agonist administration [9]. Since the inability to experience pleasure is one of the major symptom of depression [19], it may be expected that the clinical effect of AD is substantially connected with their action on reward functions.

These considerations prompted us to study the effect of imipramine (IMI), a typical tricyclic AD, on the rewarding effectiveness of different reinforcers which are known to interfere with the central DA system.

We used a place preference paradigm in which the rewarding properties of primary reinforcers, measured in terms of their ability to produce conditioned reinforcement, are estimated in the absence of a primary reward. It has been postulated that this rate-independent model can provide more reliable evidence for the efficacy of reinforcement than other methods employing measurement of the rate of operant responding under the unconditioned stimulus delivery [11,16]. In the experiments two reinforcers were used: the pharmacological agent apomorphine (APO), and the natural reinforcer—food given to hungry rats. The pairing either of them with distinctive environmental stimuli resulted in an ac-

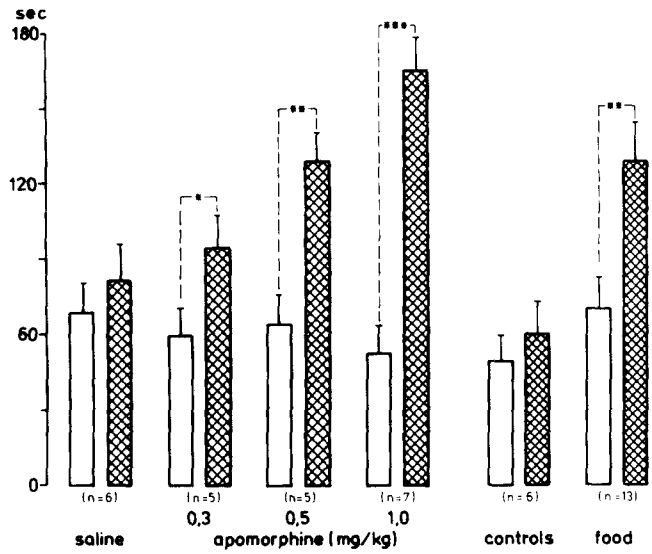

FIG. 1. Place preference conditioning induced by systemic injection of different doses of apomorphine and presentation of food to hungry rats. Bars represent means (±SEM) of the time spent in the nonpreferred compartment during pre- (open bars) and postconditioning (double-slashed bars) test. *$p<0.05$, **$p<0.01$, ***$p<0.001$.

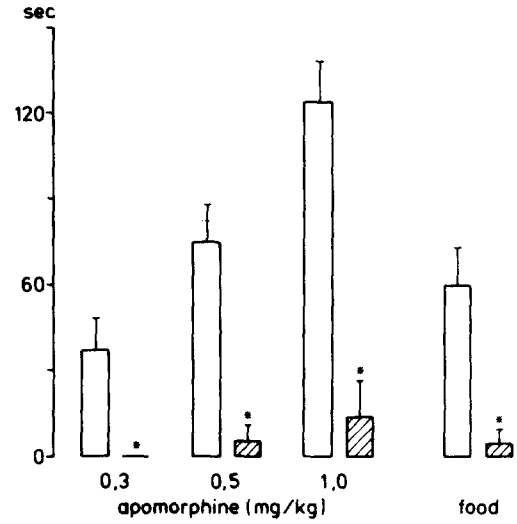

FIG. 2. Effect of short-term administration of IMI (slashed bars) and saline (open bars) on the APO- and food-induced place preference. Data represent means (±SEM) of the difference in the time spent in the compartment paired with APO or food between the pre- and postconditioning tests. Each group consisted of 5–6 animals. *$p<0.001$.

quired preference for those stimuli; moreover, it was found that this conditioned effect is mediated by the DA system [27,28].

METHOD

Animals

The subjects were male Wistar rats weighing 220–250 g. The animals were housed in groups (7 animals per cage) on a reversed 12-hour light/dark cycle (light at 08.00 p.m.). The room temperature was maintained at 22±2°C. Water and food (LSM, Bacutil) were available ad lib, except for the experiment with food as a reinforcer.

Apparatus and Procedure

The place preference apparatus was a rectangular box divided into two compartments (45×40×30 cm) connected by an exterior corridor (40×10×20 cm). Each of two compartments was distinctive in the colour of the walls covering, one having black and the other white walls. The test apparatus was equipped with a loud-speaker generating a continuous, "white", masking noise at 60 dB. The procedure consisted of 3 phases:

Phase 1. For the first 3 days the animals were allowed to explore the both compartments for 20 min per day. On day 4 each rat was placed in the corridor, and the time spent in each compartment was measured in a 10-min preconditioning test. The compartments in which the animals spent more and less time were called preferred and nonpreferred, respectively. For further phases of the experiment only rats that spent more than 6 min in the preferred compartment, less than 2 min in the nonpreferred compartment, and less than 1.5 min in the corridor were allowed.

Phase 2.—The experiment with APO as a reinforcer. The separate groups of animals were injected with different doses of APO (0.3, 0.5 and 1.0 mg/kg), and 10 min later they were individually confined in their nonpreferred compartment for 30 min. Control rats received saline and were trained in a similar way. The procedure was repeated for 3 days.

—The experiment with food as a reinforcer. Hungry rats, previously well-adapted to a limited access to food (2 hr per day), were individually confined in their nonpreferred compartment for 2 hr, where they had free access to food (LSM, Bacutil) and water. Control animals were trained likewise, but food was not presented in the conditioned compartment. The procedure was repeated for 4 days. In all the experiments with food used as unconditioned stimulus, the amount of food intake during conditioning period was measured for each animal.

Phase 3. On the day after the last session of Phase 2, each animal was placed in the corridor, and the time spent in each compartment was measured again in a 10-min postconditioning test.

During both the pre- and postconditioning tests no drug was injected, no food was available and the animals were fed prior to the test in their home cages. Thus, primary reinforcers were absent, and the increased time that the animals spent in the compartment previously associated with the drug or food in the postconditioning test relative to the preconditioning test, would index the degree of place preference conditioning produced by primary reinforcers.

IMI Administration

Each experiment was conducted on a separate group of animals, and the animals were used only once. The effect of short-term IMI treatment was investigated by administration of the drug (10 mg/kg PO) 2 hr before each session of Phase 2 (injection of APO, or presentation of food). IMI was not given before both pre- and postconditioning tests (Phase 1,3). To study the effect of long-term treatment with IMI, the

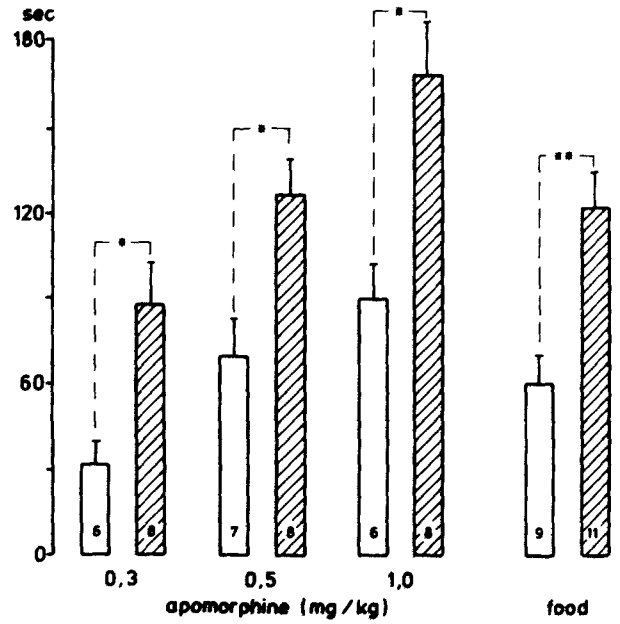

FIG. 3. Effect of long-term administration of IMI (slashed bars) and saline (open bars) on the APO- and food-induced place preference. Data represent means (±SEM) of the difference in the time spent in the compartment paired with APO or food between the pre- and postconditioning tests. The number of rats in each group is given at the bottom of columns. *$p<0.05$, **$p<0.001$.

drug was administered twice a day in a dose of 10 mg/kg PO. Successive phases of the place preference procedure began on day 9 of IMI administration, and were carried out 2 hr after the morning dose of the drug. In order to determine the effect of IMI by itself, a dose of 10 mg/kg PO of the drug was administered, and 2 hr later the animals were confined in their preferred compartment for 2 hr. The procedure was repeated for 3 days. During the pre- and postconditioning tests IMI was not given.

Drugs

The drugs used in the experiments were: apomorphine hydrochloride (Sandoz) and imipramine hydrochloride (Polfa). Both the drugs were dissolved in 0.9% sterile saline and were administered in a volume of 1 ml/kg. APO was injected subcutaneously.

Statistical Analysis

The behavioral data were analysed by an analysis of variance (ANOVA) followed by the Newman-Keuls test.

RESULTS

Figure 1 shows the degree of place preference conditioned to APO and food. In comparison with the preconditioning test, the time spent by animals in the compartment that had been previously associated with injection of different doses of APO (0.3, 0.5, and 1.0 mg/kg SC) or presentation of food was significantly prolonged in the postconditioning test, $F(1,32)=11.19$ for APO, $F(1,14)=15.79$ for food; $p<0.01$. The significant differences between the APO-treated groups indicated a dose-dependence of the place preference. The behaviour of control animals was not changed by the training procedure, $F(3,20)=1.02$; n.s.

TABLE 1

TIME (sec) ± SEM SPENT IN THE COMPARTMENT (INITIALLY PREFERRED), PAIRED WITH IMI OR SALINE ADMINISTRATION, BEFORE AND AFTER CONDITIONING

Group	n	Preconditioning Test	Postconditioning Test
Saline	7	388 ± 26	402 ± 39
IMI (10 mg/kg)	7	432 ± 34	250 ± 37*

*$p<0.01$; compared to preconditioning test.

Short-term administration of IMI prevented the development of the conditioned place preference produced by both reinforcers (Fig. 2). In the postconditioning test the animals showed no visible preference for the compartment previously paired with APO or food. In consequence, the difference between the amount of time spent in the conditioned compartment during the pre- and postconditioning tests was significantly smaller in the IMI-treated animals than in controls, $F(1,31)=18.32$ for APO, $F(1,9)=29.01$ for food; $p<0.001$.

On the other hand, long-term pretreatment with IMI enhanced the APO- and food-induced place preference conditioning (Fig. 3). The difference between the time spent in the conditioned compartment during the pre- and postconditioning tests was significantly greater in the animals treated repeatedly with IMI than in controls, $F(1,40)=11.90$ for APO, $F(1,18)=14.79$ for food; $p<0.01$. In both cases (short- and long-term treatment with IMI) no significant changes in food intake during each conditioning session were found.

Table 1 shows place aversion evoked by IMI. In the postconditioning test the animals spent significantly less time in the compartment previously associated with that drug, $F(1,12)=12.74$, $p<0.01$. The behaviour of control animals was not affected by the training procedure, $F(1,12)=2.36$; n.s.

DISCUSSION

The experiments demonstrate that the systemic injection of APO as well as the presentation of food to hungry rats evoke an approach response and produce a conditioned place preference. These results agree with some earlier reports showing that both APO and food are able to bestow reinforcing properties on the initially neutral environmental cues, which can then act as conditioned incentive stimuli in the absence of a primary reward [18,27,28].

The development of the conditioned place preference was abolished when each conditioning session was preceded by administration of single doses of IMI. Such an inhibitory effect of short-term treatment with IMI was rather surprising, since it appeared to be inconsistent with the earlier findings. In other behavioural paradigms, in which the response of animals was also initiated and maintained by appetitive stimuli (self-stimulation, operant behavior), either no effect or enhancement, rather than the decrease in the reinforcement rate after an acute treatment with AD was reported [2,22]. In the present experiment it was found that single doses of IMI used as unconditioned stimuli produced the

conditioned place aversion. Therefore it seems that the lack of positive conditioned effect in animals treated with IMI on a short-term basis was due to the cancellation of two independent but opposite effects: APO- or food-induced preference and IMI-induced aversion. Nevertheless, as no data concerning the affective properties of IMI are available, both the psychophysiological and neurochemical background of the above-mentioned effects of IMI remains unknown.

In contrast to short-term effects of IMI, the long-term pretreatment with the drug significantly enhanced the APO- and food-induced place preference. The mechanism of this effect seems to be closely related with the action of IMI on the central DA system and with the postulated role of DA receptors in the neuronal mechanism of reward [31]. In the place preference conditioning the initially neutral environmental cues gain—via classical conditioning—incentive properties of unconditioned stimuli. As a result, in the postconditioning test the animals exhibit a conditioned approach response to a degree dependent upon appetitive properties of the unconditioned stimuli [13]. As it was stated above, chronic treatment with IMI and other AD leads to hypersensitivity of postsynaptic mesolimbic DA receptors and potentiates the animal response to DA agonists [15,25]. While increasing the sensitivity of DA receptors, IMI enhances the efficacy of APO as a reinforcer and potentiates the incentive strength of the conditioned reinforcement.

There are evidences indicating that the ingestive behavior is also accompanied with changes in the functional activity of DA neurons. In particular, it was found that when hungry rats are given free access to food, a significant increase in the DA release from presynaptic terminals is observed in some limbic structures [7,8]. Therefore the above assumption concerning the effect of APO may also be true for the mechanism by which the long-term treatment with IMI enhances the conditioned preference for the compartment associated with the presentation of food. The results showing the increased conditioned response to both kinds of primary reinforcers indicate that the above-mentioned effect of long-term treatment with IMI is not exclusively related to the pharmacological activity of APO, but also appears as a result of activation of the physiological mechanism responsible for the development and incentive properties of the conditioned reinforcement. Such an activating effect of prolonged AD administration on the reward processes has been also demonstrated in the self-stimulation paradigm, as well as in rats performing under a DRL schedule of reinforcement [1, 6, 12].

It should be noted that in the present experiments primary reinforcers were associated with the initially nonpreferred compartment; thus the obtained results could be confounded by the preexisting biases in the test apparatus. However, recent studies showed the similar conditioned effect to DA agonists under the procedure in which animals did not exhibit any preference for either compartments prior to drug conditioning [17]. Moreover, the finding that the place preference conditioning can be produced by a natural reinforcer (food given to hungry rats) argues against a possible participation of a mechanism different from animal reward functions (e.g., drug-induced attenuation of the aversiveness to the nonpreferred compartment) in the above-mentioned effects.

Studies into the mechanism of action of AD have been mainly based on the analysis of their pharmacological interaction with other drugs (see [14]). However, the obtained results do not allow determination of the effect of AD on the physiological processes whose modification might be important for the antidepressive properties of these drugs. The present functional studies demonstrate an enhancing effect of long- but not short-term treatment wih IMI on the DA-mediated reward functions and, in consequence, on the rewarding value of both artificial (i.e., pharmacological) and natural reinforcers. Since the "anhedonia," perhaps due to deficits in incentive motivation, is the most important factor observed in patients suffering from depression [10], all these results seem to confirm the hypothesis about a possible disfunction of the DA system in this illness [29]. Moreover, they allow a conclusion that adaptive changes in the DA system, observed in pharmacological studies after prolonged treatment with AD, may, in fact, be of crucial importance for the clinical antidepressant properties of these drugs.

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