The effects of amphetamine, apomorphine, SKF 38393, quinpirole and bromocriptine on responding for conditioned reward in rats

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The present study investigated the effects of the dopamine (DA) D1 selective agonist, SKF 38393, and the D2 selective agonists, quinpirole and bromocriptine, on responding for conditioned reward. The nonselective DA agonist apomorphine and the indirect agonist amphetamine, were also evaluated. Male rats ($n = 302$) were tested in a procedure consisting of three distinct phases. During the pre-exposure phase the rats were exposed to an operant chamber containing two levers; one lever produced a lights-off stimulus (3 s) and the other a tone stimulus (3 s). This was followed by 4 conditioning sessions during which the levers were removed and rats received pairings of the lights-off stimulus (80 per day) and food, presented according to a variable time 45 s schedule. Two test sessions followed during which the levers were present and the number of responses made on each lever was calculated as a ratio of the number of responses made during pre-exposure. Drugs were administered prior to each test session. A saline group showed a higher ratio of responding for the lights-off stimulus than the tone stimulus, indicating that the lights-off stimulus had become a conditioned reward. Amphetamine (0.01-2.0 mg/kg) and to a lesser extent, quinpirole (0.01-5.0 mg/kg) and bromocriptine (0.05-10.0 mg/kg) dose-dependently increased responding and specifically enhanced responding on the lever producing the conditioned reward. Apomorphine (0.1-5.0 mg/kg) increased responding on both levers at higher doses but the conditioned reward effect was lost. SKF 38393 (0.1-10.0 mg/kg) appeared to impair the acquisition of responding for conditioned reward. The results were interpreted as indicating that responding for conditioned reward may be dependent on a D1 receptor-mediated reward signal.

Keywords: Amphetamine – Apomorphine – Bromocriptine – Conditioned reward – Dopamine – D1 receptors – D2 receptors – Quinpirole – SKF 38393

INTRODUCTION

There is an accumulation of evidence suggesting that drugs that influence dopamine (DA) neurotransmission can affect responding for conditioned reward. A conditioned reward is a previously neutral stimulus that has acquired rewarding properties by having been paired repeatedly with an unconditioned rewarding stimulus such as food. Skinner (1938) described this phenomenon as conditioned reinforcement. We use the term "reward" in place of "reinforcement" for theoretical reasons as outlined elsewhere (Miller *et al.*, 1990; Beninger, 1991). In one variation of the conditioned reward paradigm animals first received pairings of a stimulus (e.g. light) with a primary reward. Testing was carried out in an operant chamber containing two levers, one of which produced the reward-related stimulus. When animals responded more for the stimulus that previously was paired with reward it was concluded that the stimulus had become a conditioned reward (e.g. Stein, 1958).

There may be a DA signal associated with the presen-tation of rewarding stimuli. *Ex vivo* or *in vivo* neurochemical studies have revealed that DA release in the nucleus accumbens or caudate nucleus of rats increased when they pressed a lever for food, brain stimulation or drug reward (Blackburn *et al.*, 1986; Hernandez and Hoebel, 1988; Joseph *et al.*, 1989; Phillips *et al.*, 1989; Radhakishun *et al.*, 1991; Nakahara *et al.*, 1991). Other studies have shown increased DA release in association with the presentation of conditioned stimuli signalling food (Blackburn and Phillips, 1989; Phillips *et al.*, 1991). These results suggest that there may be a DA signal produced by unconditioned or conditioned rewarding stimuli (see Beninger, 1991).

Some studies have evaluated the effects of DA antagonists on the establishment of conditioned reward. Pimozide blocked the effect (Beninger and Phillips, 1980; Hoffman and Beninger, 1985). As pimozide was administered during the conditioning phase and the animals were subsequently tested drug-free, it is unlikely that the effects

were attributable simply to motor debilitation. These results suggested that DA participates in the establishment of a neutral stimulus as a conditioned reward.

Other studies have focused on the effects of psychomotor stimulants on responding for conditioned reward. Responding of rats or pigeons for conditioned reward was specifically enhanced by DA agonists such as pipradrol, amphetamine or cocaine (Hill, 1970; Robbins, 1975, 1976, 1978; Robbins and Koob, 1978; Beninger et al., 1980, 1981; Robbins et al., 1983; Mazurski and Beninger, 1986; Files et al., 1989; Cohen and Branch, 1991; but see Cohen, 1991). Similar effects of amphetamine were seen when it was microinjected into the accumbens but not the dorsal caudate nucleus (Taylor and Robbins, 1984, 1986; Kelley and Delfs, 1991a, b). Animals injected with pipradrol or microinjected with amphetamine into the nucleus accumbens did not increase responding for a stimulus that was randomly correlated with water reward during conditioning (Robbins, 1976; Taylor and Robbins, 1984). This indicated that the conditioned reward effect depended crucially on the positive contingency between the conditioned and unconditioned rewarding stimuli and that pipradol or amphetamine acted on responding for the conditioned reward rather than for stimulus change. These results showed that DA may participate in the ability of a conditioned reward to control responding (cf. Phillips and Fibiger, 1990).

Of particular interest for the present study are those experiments reporting differential effects of amphetamine and apomorphine in a conditioned reward paradigm (Robbins et al., 1983; Mazurski and Beninger, 1986). Amphetamine produced its rate-increasing effects on the lever that was associated with conditioned reward; in contrast, apomorphine produced rate-increasing effects on both levers.

The differential effects of amphetamine and apomorphine may be related to their different mechanisms of action. Amphetamine acts indirectly by enhancing the neurogenic release of DA and blocking re-uptake (Scheel-Kruger, 1971; Westerink, 1979) whereas apomorphine directly stimulates DA receptors (Colpaert et al., 1976). With these differences in mind, Herberg et al. (1976) reasoned that indirectly-acting stimulants might enhance the reward signal by facilitating the neuronal activity generated by the rewarding stimulus. Directly-acting stimulants would excite postsynaptic receptors in a relatively tonic fashion leading to a possible masking of the reward signal generated by the rewarding stimulus. This might explain the observation that amphetamine enhanced responding for conditioned reward whereas apomorphine led to indiscriminate responding (see Robbins et al., 1983).

To clarify further the role of DA in the mediation of reward, the present experiments examined the effects of DA receptor-subtype-specific agonists on responding for

conditioned reward. DA receptors can be divided into at least two sub-categories depending on their relation to the enzyme adenylate cyclase: D1 receptors stimulate, whereas D2 receptors either do not stimulate or inhibit the stimulation of this enzyme (Kebabian and Calne, 1979). It is possible that the hypothetical masking of the reward signal by apomorphine takes place at one or both of these receptors.

One recent report presents results directly relevant to this question. Nakajima and O'Regan (1991) examined the effects of D1 and D2 agonists on responding for brain stimulation reward. They found that D2 agonists shifted the rate-frequency curve to the left, an effect like that previously seen with amphetamine (Gallistel and Karras, 1984). A D1 agonist had little effect at low doses whereas higher doses led to a cessation of responding, reminiscent of the effects of apomorphine (Herberg et al., 1976). These results suggest that the apparent masking effects of apomorphine on the putative reward signal may take place at the D1 receptor. This finding supports the hypothesis that it may be the D1 receptor that mediates the effects of reward on behaviour (cf. Beninger et al., 1989; Miller et al., 1990; Beninger, 1991, 1992).

If the D1 hypothesis is correct it might be expected that D2 receptor agonists would lead to increased responding in the conditioned reward paradigm but would leave the reward signal intact. D1 agonists, on the other hand, might mask the reward signal. To test this hypothesis the present experiments investigated the effects of a range of doses of receptor-subtype-specific DA agonists on responding for conditioned reward. For the purpose of comparison, the effects of apomorphine and amphetamine also were evaluated.

METHOD

Subjects

Treatment of the rats in the present study was in accordance with the Animals for Research Act, the Guidelines of the Canadian Council on Animal Care and relevant University policy and was approved by the Queen's University Animal Care Committee.

Male Wistar rats ($n = 302$), obtained from Charles River Canada, weighing between 225 and 275 g (free-feeding) were individually housed in a temperature controlled environment ($21°C$) on a 12 h light-dark cycle (lights on at 06.00 h). Rats were habituated to the housing environment for approximately 1 week and their weights increased by 25-40 g. Weights were then reduced to 80% of these values, for the 11 day duration of the experiment, through daily feedings with measured rations.

Apparatus
The experimental environments consisted of four simi-

larly constructed operant chambers. The dimensions were 29 cm in length, 23 cm in width and 19 cm in height. The chambers were constructed of aluminum sides and plexiglass tops and doors. The floors consisted of aluminum grids. Each chamber was placed in a ventilated sound-attenuating box. Each 29 cm wall of each chamber contained a 7.5 cm by 3.5 cm removable lever, depression of which required a force of approximately 0.09 N. At the center of the 23 cm wall was located a 2.0 by 4.0 cm feeder cup at a height of 2.5 cm from the floor. An illuminated 2 W light bulb was positioned on each side (8.5 cm apart) of the feeder cup at a height of 10 cm from the floor. Each chamber also contained a 4.9 kHz tone generator positioned at 14 cm from the floor between the two light bulbs and at the center of the 23 cm wall. The tone generator was adjusted to deliver a tone 10 dB above the background noise level.

Procedure

Each group was exposed for a total of 11 consecutive days to an experimental design that consisted of three distinct phases referred to as the pre-exposure, conditioning and test phase.

The pre-exposure phase consisted of a total of five 40-min sessions held at approximately the same time on each of the first 5 days. Two levers were present. One lever produced a tone stimulus and the other a lights-off stimulus. Both stimuli lasted 3 s. Two of the chambers had the tone-producing lever on the right wall and the lights-off-producing lever on the left wall; the relationship between lever side and stimulus was reversed for the other two chambers. The number of responses on each lever was measured for each pre-exposure session.

The conditioning phase consisted of 60 min sessions held on each of the next 4 days. During conditioning both levers were removed from the operant chamber and the rats were exposed to 80 presentations of the 3 s lights-off stimulus according to a random time 45 s schedule, in which the mean time between lights-off stimulus presentations was 45 s (range 5-90 s). During the first conditioning session each lights-off stimulus presentation was terminated with the delivery of one 45 mg food pellet (Bioserv). During the remaining three conditioning sessions food delivery occurred following a random 33% of the lights-off stimulus presentations. This procedure was employed as Knott and Clayton (1966) observed that partial pairing resulted in a greater magnitude of conditioned reward than continuous pairing.

The test phase consisted of two 40 min sessions held on the two final days. The levers were again present in the operant chambers and the number of responses on each lever was measured. Conditioned reward was observed as a relative increase in the number of responses on the lights-

off stimulus lever in the test phase compared to the pre-exposure phase.

A total of 37 groups was tested. One group ($n = 16$) received saline (1 ml/kg) 5 min prior to each test session. Six groups ($n = 6$-8) received amphetamine in doses of 0.01, 0.10, 0.25, 0.50, 1.00 and 2.00 mg/kg, i.p., 5 min prior to each test session. Five groups ($n = 6$-14) received apomorphine in doses of 0.1, 0.5, 1.0, 2.0 and 5.0 mg/kg, i.p., 5 min prior to each test session. Five groups ($n = 8$) received the D1 agonist, SKF 38393, in doses of 0.1, 0.5, 1.0, 5.0 and 10.0 mg/kg, i.p., 5 min before each test session. Nine groups ($n = 8$) received the D2 agonist, quinpirole, in doses of 0.01, 0.025, 0.05, 0.10, 0.25, 0.50, 1.0, 2.5 and 5.0 mg/kg, i.p., 5 min before each test session. Seven groups ($n = 6$-8) received the D2 agonist, bromocriptine, in doses of 0.05, 0.10, 0.50, 1.0, 2.5, 5.0 and 10.0 mg/kg, i.p., 60 min prior to each test session.

The remaining four groups ($n = 7$-8) were added to evaluate possible alternative interpretations of the results with amphetamine. Two groups received pairings of the tone and food during conditioning sessions. One group received saline and the other amphetamine (0.5 mg/kg, i.p.) prior to each test session; the 0.5 mg/kg dose of amphetamine was chosen because it produced the largest enhancement in responding for conditioned reward in the amphetamine experiment (see Results). The final two groups received neither food nor lights-off nor tone stimuli during conditioning. Prior to test sessions, one of these groups received saline and the other, amphetamine (0.5 mg/kg, i.p.).

Drugs

(+)-Amphetamine sulphate (Smith, Kline and French Canada Inc.) and quinpirole (Eli Lilly and Co.) were dissolved in saline and injected in a concentration of 1 ml/kg of body weight. SKF 38393 (Research Biochemicals Inc.) was dissolved in distilled water and injected in a concentration of 1 ml/kg except for the highest dose (10.0 mg/kg) which was injected in a concentration of 2 ml/kg. Apomorphine (Sigma) was dissolved with ascorbic acid (1 mg/ml) in distilled water and injected in a volume of 1 ml/kg. Bromocriptine (Research Biochemicals Inc.) was suspended in one drop of polyoxyethylene sorbitan mono-oleate (Tween 80) and added to distilled water to achieve an injection volume of 1.0 ml/kg. All drugs were prepared daily immediately prior to injection.

Data analyses

The data within the last 30 min provided the most stable estimate of pre-conditioning response rates. In previous studies (Hoffman and Beninger, 1985) the number of responses in each 10 min segment of pre-exposure sessions was analyzed and it was found that rates were higher in the first 10 min but did not differ significantly for the remain-

ing 10 min periods. Therefore, only data from the last 30 min of the session were used in the analyses of the present results. The number of responses made on each lever during the last 30 min of the five pre-exposure sessions was averaged for each rat. The number of responses made on each lever during the last 30 min of each test session was averaged for each rat. Finally, the number of responses on each lever in each test session was divided by the number of responses on that lever in the pre-exposure phase. These ratios were square root transformed [adding 1.0 to each value entering into the ratio to reduce the influence of numerically small numbers (see Winer, 1971)] to normalize their distribution for the purposes of analyses (Keppel, 1982). Thus, the data consisted of two numbers for each rat.

To evaluate the conditioned reward effect in the saline group, a one-way analysis of variance (ANOVA) compared the ratios for each lever. A significantly higher ratio of responding on the lights-off lever than on the tone lever was taken as evidence that conditioned reward had occurred. The results for groups receiving each drug were subjected to two-way ANOVAs with repeated measures on the lever factor. When only a dose effect was seen, multiple comparisons, using the Newmann Keuls procedure, determined the source of this effect. When a lever effect or a lever by dose interaction was seen, the data were re-analyzed, this time including the saline group. The error term for the interaction from this ANOVA was then used to make interaction comparisons between the saline group and each drug dose (Keppel, 1982). When the ratio for the lights-off lever was greater than the ratio for the tone lever for a drug dose and there was a significant interaction of lever and dose in the comparison with saline, it was concluded that the dose enhanced responding for conditioned reward.

Finally, the ratios for each of the four groups included to evaluate possible alternative interpretations of the results with amphetamine were analyzed. The pair of groups receiving tone-food pairings in the conditioning phase, and the pair of groups receiving no food, tone or lights-off stimuli in the pairing phase were each subjected to a two-way ANOVA with treatment (saline or amphetamine) and levers as the factors analyzed. A significant interaction was further analyzed by tests of simple main effects of lever in each group.

RESULTS

Responding on each lever, in the test phase, for the saline, amphetamine and apomorphine groups is shown in Fig. 1. The saline group (upper panel to the left) showed a small but reliable conditioned reward effect (i.e. more responding on the lights-off lever). Groups treated with amphetamine showed a dose-related increase in responding and
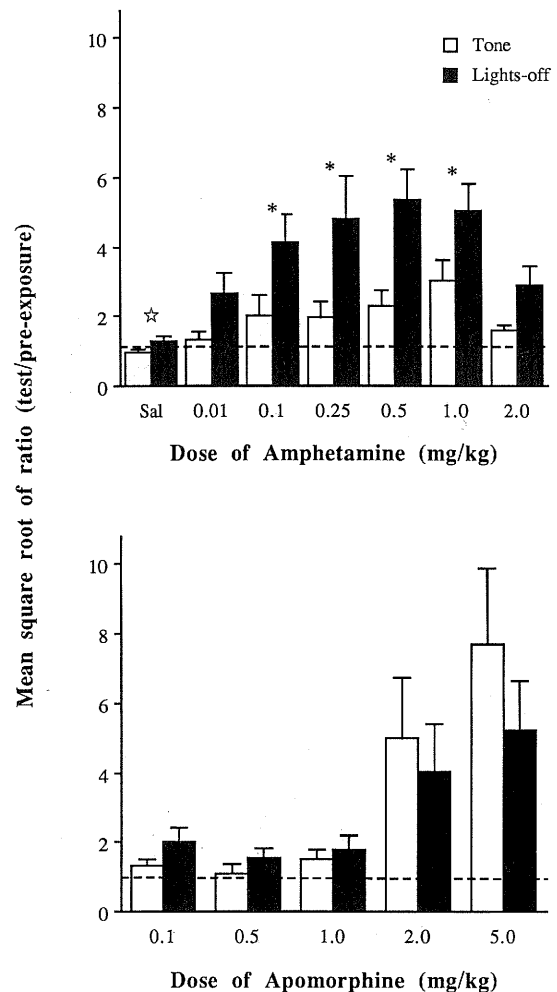

FIG. 1. Mean square roots of ratios of test phase responding relative to pre-exposure responding on each lever for the saline (Sal) group (also shown in Fig. 2) and groups receiving amphetamine or apomorphine. Vertical lines represent standard errors of the mean. The horizontal line at a ratio value of 1.0 indicates no change in number of responses from pre-exposure to test. Lights-off was the conditioned stimulus; tone was the neutral stimulus. Saline, amphetamine or apomorphine were administered i.p., 5 min before test sessions. ☆ Significant ($p < 0.01$) conditioned reward effect in the saline group; *significant enhancement of conditioned reward effect compared to saline.

pressed the lights-off lever more. In contrast, groups treated with apomorphine, although showing a dose-related increase in responding, pressed more on both the lights-off and tone levers and conditioned reward was not seen.

This description of the results was supported by the statistical analyses of the ratio data. Thus, the saline group showed a higher ratio of responding on the lights-off lever than on the tone lever [$F(1,15) = 7.80$, $p < 0.01$]. Groups treated with amphetamine showed significant dose [$F(1,5) = 2.75$, $p < 0.05$], and lever effects [$F(1,5) = 46.09$, $p < 0.001$]. The lever effect revealed that groups treated
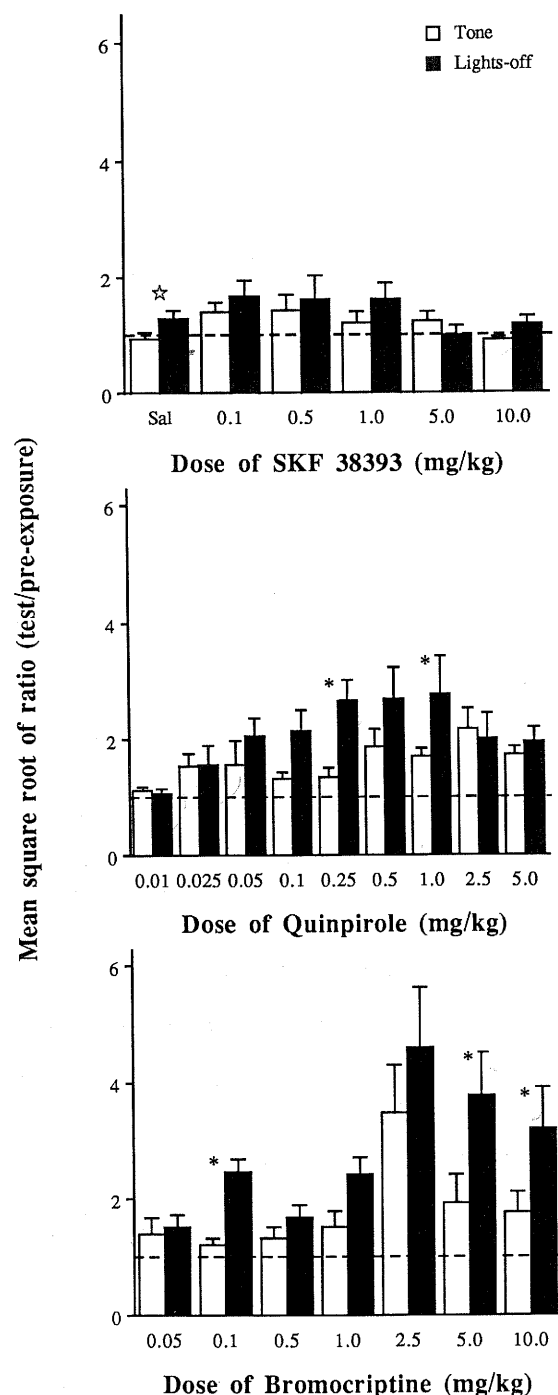
FIG. 2. Mean square roots of ratios of test phase responding relative to pre-exposure responding on each lever for groups receiving saline (Sal; also shown in Fig. 1), SKF 38393, quinpirole or bromocriptine. Vertical lines represent standard errors of the mean. The horizontal line at a ratio value of 1.0 indicates no change in number of responses from pre-exposure to test. Lights-off was the conditioned stimulus; tone was the neutral stimulus. SKF 38393 or quinpirole were administered i.p., 5 min before test sessions; bromocriptine was administered i.p., 60 min before test sessions. ☆ Significant ($p < 0.01$) conditioned reward effect in the saline group; *significant enhancement of conditioned reward effect compared to saline.

with amphetamine showed a greater increase in pressing the lights-off lever than the tone lever from pre-exposure to test, a conditioned reward effect. To determine if amphetamine produced an enhancement in comparison to saline, the ANOVA for the amphetamine groups was carried out again, this time incuding the saline group. The analysis revealed a significant interaction of lever and dose [$F_{(6,52)} = 3.16$, $p < 0.01$]. Interaction comparisons revealed that doses of 0.1, 0.25, 0.5 and 1.0 mg/kg of amphetamine led to a significant enhancement of the conditioned reward effect. Finally, groups treated with apomorphine showed only a significant dose effect [$F_{(4,37)} = 6.59$, $p < 0.001$]. Multiple comparisons revealed that doses of 2.0 and 5.0 mg/kg led to significantly more responding than the three lower doses. Apparently, apomorphine produced a dose-dependent increase in responding but the conditioned reward effect was not seen.

Responding on each lever, in the test phase, for SKF 38393, quinpirole and bromocriptine groups is shown in Fig. 2. Groups treated with SKF 38393 showed little or no evidence of conditioned reward depending on the dose. Groups treated with quinpirole showed a small dose effect and higher ratios of pressing the lights-off lever at the middle of the dose range. Groups treated with bromocriptine showed a dose effect and a conditioned reward effect at several doses.

This description of the data is supported by the results of statistical analyses as follows. Analysis of the SKF 38393 doses revealed no significant effects. The lack of a lever effect suggests that treatment with SKF 38393 may have led to a failure to see conditioned reward. Groups treated with quinpirole showed a significant lever effect [$F_{(1,8)} = 15.06$, $p < 0.001$], and the effects of dose [$F_{(8,63)} = 1.98$, $p < 0.06$], and interaction [$F_{(8,63)} = 1.88$, $p < 0.08$], approached significance. Thus, groups treated with quinpirole tended to show a conditioned reward effect. When the saline group was included in the ANOVA for the quinpirole groups, the interaction was significant [$F_{(9,78)} = 2.01$, $p < 0.05$]. Interaction comparisons revealed that quinpirole doses of 0.25 and 1.0 mg/kg produced an enhancement of the conditioned reward effect. Groups treated with bromocriptine, like those treated with the D2 agonist quinpirole, showed a significant lever effect [$F_{(1,44)} = 33.07$, $p < 0.001$]. The dose effect also was significant [$F_{(6,44)} = 4.43$, $p < 0.001$]. When saline was included in the ANOVA, the interaction reached significance [$F_{(7,59)} = 2.47$, $p < 0.05$]. Interaction comparisons with the saline group showed that bromocriptine doses of 0.1, 5.0 and 10.0 mg/kg produced a significant enhancement of the conditioned reward effect.

Responding on each lever in the test phase for the groups receiving no food, lights-off or tone stimuli during conditioning and the groups receiving tone-food pairings
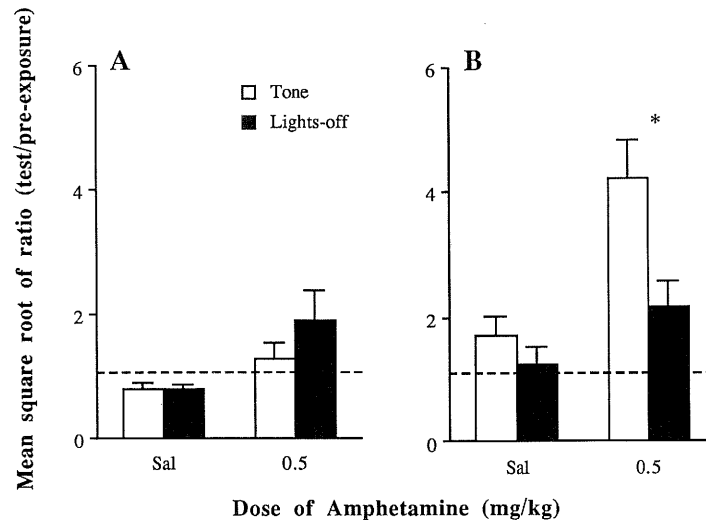
FIG. 3. Mean square roots of ratios of test phase responding relative to pre-exposure responding on each lever for groups receiving either no tone, no lights-off and no food stimuli (A) or receiving tone-food pairings (B) during conditioning and saline (Sal) or amphetamine (0.5 mg/kg) in the test. Vertical lines represent standard errors of the mean. The horizontal line at a ratio of 1.0 indicates no change in number of responses from pre-exposure to test. Saline or amphetamine were administered i.p., 5 min before test sessions. *Significant enhancement of conditioned reward effect compared to saline.

during conditioning are shown in Fig. 3. The group receiving no food, lights-off or tone stimuli in the pairing phase and saline in the test showed little change in responding on either lever. Amphetamine led to an increase in responding on both levers with a greater increase on the lights-off lever. The groups receiving tone-food pairings in the conditioning phase and saline in the test showed a preference for the tone lever in the test. The group receiving amphetamine in the test following tone-food pairings in the conditioning phase showed an enhancement of the conditioned reward effect.

Statistical analyses supported this description of the data. The ANOVA comparing ratios on the two levers for the groups receiving no food, tone or lights-off stimuli in the conditioning phase revealed significant effects of treatment $[F(1,12)=4.74, p<0.05]$, lever $[F(1,12)=4.55, p<0.05]$, and lever by treatment interaction $[F(1,12)=4.56, p<0.05]$. The significant interaction seemed to be due to greater responding on the lights-off lever in the amphetamine group. However, tests of simple main effects of lever for each group failed to reveal significant effects in either of them. The ANOVA comparing ratios on the two levers for the groups receiving tone-food pairings revealed significant lever $[F(1,14)=19.29, p<0.001]$, treatment $[F(1,14)=10.73, p<0.01]$, and interaction effects $[F(1,14)=7.44, p<0.05]$. Tests of simple main effects of lever for each treatment revealed a significant effect in the amphetamine group

$[F(1,7)=19.53, p<0.001]$. Thus, pairing the tone with food led to a small nonsignificant conditioned reward effect that was significantly enhanced by amphetamine.

DISCUSSION

For the saline group, pairing of the lights-off stimulus with food resulted in the lights-off stimulus becoming a conditioned reward. Previous control studies in which the conditioned stimulus was negatively correlated with reward (Hoffman and Beninger, 1985; Beninger and Phillips, 1980) failed to observe a similar effect revealing the importance of the positive contingency between the conditioned stimulus and the food pellets. These results are consistent with previous findings from this (Hoffman and Beninger, 1985; Mazurski and Beninger, 1986) and other laboratories (Skinner, 1938; Stein, 1958; Hill, 1970; Robbins, 1978; Robbins and Koob, 1978; Files et al., 1989).

Rats given amphetamine showed dose-dependent increases in responding specifically on the lever producing the lights-off stimulus. In addition, most of the amphetamine groups showed larger conditioned reward effects than the saline group. These results are consistent with those obtained by Robbins et al. (1983) and Mazurski and Beninger (1986) and indicate that amphetamine produces an enhancement of responding for conditioned reward. It is interesting to note that Cohen and Branch (1991), studying the responding of pigeons on second order schedules

of conditioned reward, recently reported that amphet-amine enhanced responding during a period of extinction. These findings are in excellent agreement with those reported here.

An alternative explanation for the effects of amphet-amine would be that it alters sensitivity to light (Isaac, 1971; Goetsch and Isaac, 1983) possibly leading to enhanced responding for the lights-off stimulus. We tested this hypothesis in two ways. First, groups received no food, tones or lights-off stimuli in the conditioning phase and either saline or amphetamine (0.5 mg/kg) in the test. Amphetamine produced a small relative increase in press-ing the lights-off lever but it was only a fraction of the size of the increase seen in animals that had received 0.5 mg/kg of amphetamine following pairings of the lights-off stim-uli with food. Second, animals receiving amphetamine in the test following tone-food pairings showed a significant enhancement of the small (nonsignificant) conditioned reward effect produced by tone-food pairings in a group treated with saline in the test. We have previously observed better conditioning with the lights-off than the tone stimulus (unpublished results). Whether this is related to the specific frequency and intensity of the tone used here or to a difference in the efficacy of auditory ver-sus visual cues to act as conditioned rewards awaits further study. However, having observed this difference we car-ried out the current studies using only the lights-off stimu-lus as a conditioned reward. The observation that amphetamine significantly enhanced responding for the tone when it had been paired with food shows that the effects of amphetamine are not specific to the lights-off stimulus. The observation that the effects of amphetamine on responding on the lights-off lever were relatively small in animals receiving no conditioning similarly shows that the effects of amphetamine are not simply to augment responding for lights-off. These two control experiments together provide strong support for the conclusion that amphetamine enhances responding for conditioned reward.

Groups receiving apomorphine failed to exhibit increases in responding specifically for the lights-off stimulus suggesting that apomorphine impaired respond-ing for conditioned reward. These results are similar to those obtained by Robbins et al. (1983) and Mazurski and Beninger (1986). The two largest doses of apomorphine resulted in large increases in responding on both levers. These findings show that apomorphine has a general stimulant effect and are consistent with the results of pre-vious investigations of the locomotor effects of apo-morphine (for a detailed review see Ungerstedt, 1979).

Groups treated with SKF 38393 failed to show a reliable increase in responding for the lights-off stimulus. Thus, the D1 agonist appeared to impair the conditioned reward effect.

Animals given quinpirole or bromocriptine displayed a pattern of responding somewhat similar to those adminis-tered amphetamine. Both drugs dose-dependently increased responding. In addition, groups receiving the mid-dose range of quinpirole and the higher doses of bro-mocriptine showed conditioned reward effects larger than that in the saline group. Thus, the D2 agonists at some doses enhances responding for conditioned reward.

Drugs acting on dopaminergic systems have been shown to produce hyperactivity and various forms of stereotyped behaviour (Ungerstedt, 1979; Waddington and O'Boyle, 1989). It might be argued that apomorphine, amphetamine, quinpirole and bromocriptine produced increases in motor activity leading to increases in bar pressing. However, amphetamine, and to a lesser extent quinpirole and bromocriptine, produced increases in responding specifically on the conditioned reward lever whereas apomorphine produced an increase in responding on both levers. If an increase in DA activity simply enhanced bar pressing then these drugs should have pro-duced similar patterns of responding. Thus, a general motor stimulant effect of dopaminergic agonists cannot explain the present results.

In summary, amphetamine, quinpirole and bromocrip-tine enhanced responding for conditioned reward. In con-trast, SKF 38393 appeared to impair the conditioned reward effect. Finally, apomorphine impaired responding for the conditioned reward across a wide dose range and had a strong general stimulant effect at the higher doses. The differential effects of these DA agents may be under-stood with reference to their different mechanisms of action.

Amphetamine and apomorphine are DA agents that are not specific to either D1 or D2 receptors. Although both enhance DA neurotransmission they do so by different means. Amphetamine increases the neurogenic release and blocks re-uptake of DA (Scheel-Kruger, 1971; West-erink, 1979), intensifying DA action in active synapses. Alternatively, apomorphine directly stimulates DA recep-tors (Colpaert et al., 1976), providing relatively tonic stimulation. There is good evidence that DA is released when animals are presented with a primary or conditioned reward (Blackburn et al., 1986; Hernandez and Hoebel, 1988; Blackburn and Phillips, 1989; Joseph et al., 1989; Phillips et al., 1989, 1991; Radhakishun et al., 1991; Nakahara et al., 1991). The different mechanisms of action of amphetamine and apomorphine might influence this reward signal differently. Thus, indirect stimulants might enhance the reward signal by selectively facilitating the neuronal activity generated by the rewarding stimulus; direct stimulants might mask the reward signal by exciting postsynaptic receptors in a relatively tonic fashion. From this point of view, amphetamine, an indirect-acting stimu-lant, dose-dependently enhanced the ability of the con-

ditioned reward to control responding whereas apomorphine, a direct-acting stimulant, masked the ability of the conditioned reward to control responding (see Herberg et al., 1976; Robbins et al., 1983).

The putative masking of the reward signal by apomorphine may occur at either or both D1 and D2 receptor sites. The present findings are in good agreement with those of Nakajima and O'Regan (1991) and tend to suggest that this masking effect takes place at the D1 receptor. The observation of enhanced conditioned reward by D2 agonists would appear to rule out the possibility that the lack of a conditioned reward effect observed with apomorphine was due to its stimulation of D2 receptors. Moreover, stimulation of D1 receptors appeared to lead to a weakening of the ability of a conditioned reward to control responding. The results suggest that stimulation of the D1 receptor in association with reward is necessary for reward-related learning.

It has been suggested that the activation of DA neurons by the presentation of rewarding stimuli leads to incentive learning; it was further speculated that this learning may be mediated by a heterosynaptic mechanism involving stimulation of D1 receptors (Beninger, 1983). The present results provide indirect support for the D1 hypothesis as do the results of a number of recent studies showing that the D1 antagonist, SCH 23390, block reward-related learning (see Beninger et al., 1989; Miller et al., 1990; Beninger, 1991, 1992).

The conclusion of the present paper is that it is the stimulation of D1 receptors by DA, putatively released when a conditioned reward is presented, that leads to incentive learning, i.e. enhanced ability of the conditioned reward-producing lever to elicit approach and pressing responses. Motor activity is enhanced by D2 agonists but, at least at moderate doses of D2 agonists, putative DA release upon presentation of conditioned reward may still lead to the stimulation of D1 receptors and incentive learning. The reward signal at D1 receptors may be masked by apomorphine or SKF 38393; because apomorphine also stimulates D2 receptors, activity is enhanced but the control of responding by the conditioned reward may be lost. These considerations would lead to the prediction that the enhancement of responding for conditioned reward produced by amphetamine or D2 agonists would be blocked by a D1 antagonist. The results of recent studies support this prediction (Ranaldi and Beninger, in preparation).

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