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Role of Dopamine in *d*-Amphetamine-induced Discriminative Responding[1]

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BENG T. HO AND JEN-TZAW HUANG. *Role of dopamine in d-amphetamine-induced discriminative responding.* PHARMAC. BIOCHEM. BEHAV. 3(6) 1085–1092, 1975. – In two-lever operant chambers, rats were trained in a food-reinforced discrimination task. Reward was contingent upon correct lever choices to the induced differential cue conditions of d-amphetamine (0.8 mg/kg) or saline throughout training on a differential reinforcement of low response rate (DRL-15 sec) schedule. Upon acquisition of discriminative response control the animals were pretreated with various neurochemical agents. Pretreatment with atropine, phentolamine, propranolol, methysergide or cinanserin did not block production of d-amphetamine lever responding. Nicotine and oxotremorine did not produce the amphetamine-like cueing effect. However, pimozide blocked the ability of animals to discriminate d-amphetamine and L-DOPA, in combination with Ro 4-4602 with or without amantadine, generated amphetamine-like responses. These results indicate a role of dopamine in the production of the amphetamine state and from the failure of apomorphine to exhibit the stimulus property of d-amphetamine and the antagonism by α-methyltyrosine of the d-amphetamine responding it is further suggested that this amphetamine state is produced via the releasing of newly synthesized dopamine.

d-Amphetamine-discriminative stimulus property Neurochemical agents for monoaminergic systems Dopamine

IT has been demonstrated that centrally acting stimulants such as nicotine [35, 42, 45] and amphetamine [21, 46, 50] can serve as internal discriminative stimuli in producing an interoceptive cue in animals. Under such a condition, the animal can be trained to make responses for reward by choosing between either of two levers in an operant chamber or the arms of a T-maze according to whether the drug or saline was administered. The response control by the drug in the operant chamber is achieved when the animal, after acquiring saline versus drug discrimination, responds more than 80 percent on the drug lever during an extinction test in which the reinforcement by food is discontinued. Drugs which exhibit discriminative response control by this paradigm include amphetamine [21, 26, 28, 30, 38, 50], alcohol [22,31], atropine, chlordiazepoxide, chlorpromazine, pentobarbital, psilocybin [22], lysergic acid diethylamide [25], mescaline [9, 10, 25], and β-phenylethylamine in conbination with iproniazid [29].

It has been reported that the amphetamine-induced discriminative responding is of central origin [38,39]. The present study was undertaken to investigate the possibility that this discriminative behavior in animals can be associated with central neuroamines like other actions of amphetamine. Specifically, it was hoped that the utilization of various neurochemical agents unique for cholinergic, serotonergic, adrenergic and dopaminergic systems would aid in determining if a particular amine is responsible for the establishment of amphetamine state responding.

METHOD

Animals

Thirty male Sprague-Dawley rats (350–450 g), purchased from Horton Labs, Oakland, California, were used in the study. They were housed individually with access to water ad lib. but were maintained at 85 percent of their expected free-feeding weights by adjusted feedings.

Drugs

Methysergide maleate was kindly supplied by Sandoz Pharmaceuticals, N. J., phentolamine hydrochloride by Ciba Pharmaceutical Co., N. J., pimozide by Professor Paul Janssen of Janssen Pharmaceutical Research Laboratoria, Belgium, chlorpromazine hydrochloride by Smith Kline and French Laboratories, Pa., apomorphine hydrochloride by

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Merck Chemical Division, Merck and Co., Inc., N. J., and Ro 4-4602 by Hoffman-La-Roche, Inc., N. J. Atropine sulfate, oxotremorine sesquifumarate, nicotine sulfate, cinanserin hydrochloride, propranolol hydrochloride, L-(-)-DOPA, amantadine hydrochloride, DL-α-methyl-*p*-tyrosine methyl ester hydrochloride and d-amphetamine sulfate were purchased from commercial sources.

Apparatus

Five operant chambers (Scientific Prototype, Model A-100) each equipped with 2 operant levers (Scientific Prototype, Model PLS-100) were used for behavioral training and testing. Each operant chamber was enclosed in a sound attenuating chamber (Scientific Prototype, Model SPC-300) equipped with a fan to circulate fresh air and with a 7 W house light to provide illumination. Reinforcement consisted of single 45 mg Noyes pellets (standard formula). All behavioral contingencies and data collection were controlled by programming equipment (Grason-Stadler 1200 series). Cumulative recorders (Gerbrands, Model G-3) were used during extinction test sessions.

Procedure

Preliminary testing. Animals were allowed to become familiar with the operant chamber and lever responding, and were then trained for 4 days on a daily 30 min continuous reinforcement (CRF) schedule to respond on each lever. Differential reinforcement of low response rate (DRL) schedule was then introduced. Daily 30 min sessions were carried out for 2 days on each lever under DRL 5 sec schedule, followed by 2 days on DRL 10 sec and DRL 15 sec schedules. Thereafter, the animals were trained throughout all experiments.

Discriminative training. After the animals had stabilized on the 4 day pretraining under the DRL 15 sec schedule, the discriminative training began. Fifteen min prior to each 30 min training period, each animal was injected intraperitoneally with either d-amphetamine sulfate (0.8 mg/kg) in saline, or saline (1 ml/kg) according to the test day: on the day following amphetamine administration only responses made on the left lever were reinforced, while on the day when saline was given, reinforcement was contingent on pressing only the right lever. The sequence of weekly 4 day injections of drug and saline was a counter balance order of all the possible combinations with 2 days on the left (amphetamine) lever and 2 days on the right (saline) lever, and no more than 2 consecutive sessions utilized either drug or saline. After 32 training sessions all animals' responses were more than 80 percent correct on the appropriate lever.

Extinction test. On the fifth day of a week following the 4 day discriminative training, the animals were injected with d-amphetamine or saline 15 min prior to being placed in the operant chambers with the reinforcement delivery disconnected (extinction). The degree of discrimination between d-amphetamine and saline was reflected in the percentage of responses made on the lever appropriate to the state of the animal during 10 min testing in the absence of reinforcement feedback. After 6–8 extinction test sessions on a given Friday the discriminative control was established since all animals responded more than 80 percent on the correct lever. The accuracy of lever responses was maintained by training the animals prior to each extinction test with d-amphetamine and saline for food reinforcement.

Test trails with neurochemical agents. Extinction sessions were performed on the fifth day of every week using various agents known to affect monoaminergic nervous systems to determine their effects on the discriminative stimulus property of d-amphetamine (Table 1). Two categories of agents were given intraperitoneally to the trained animals: first antagonists and then agonists. Animals were randomly divided into groups of 5 for receiving each drug. Most of the animals were used more than once but not for the same generalization test. The dosage and time course utilized for each drug were those reported in the cited literature when the biological effect on specific monoamines reportedly was optimal.

(a) Antagonists. Antagonists were used to affect d-amphetamine-induced discriminative responding to define the role of monoamines in the discriminative stimulus property of d-amphetamine. Animals were pretreated at the designated time with these antagonists before receiving 0.8 mg/kg of d-amphetamine or saline. Extinction tests were carried out 15 min after the amphetamine or saline injection for 10 min periods. The percent of total responses on the amphetamine lever resulting from the effect of the antagonist was then calculated.

The cholinergic antagonist, atropine sulfate in saline, was injected at doses of 1 or 2 mg/kg followed 15 min later by d-amphetamine sulfate in saline. Schechter and Rosecrans [43] used atropine (0.5 mg/kg) 10 min prior to arecoline to block the discriminative stimulus property of arecoline which was tested 10 min after arecoline injection.

The serotonergic antagonists, methysergide maleate (4.5 mg/kg) and cinanserin hydrochloride (15 mg/kg), were dissolved in saline and injected 15 min prior to the amphetamine or saline. Winter [54] tested 1–6 mg/kg of methysergide in a variable interval 30 second schedule for the antagonism of rate-depressant effect of 4 mg/kg of N,N-dimethyltryptamine (DMT), which is thought to act on central tryptamine receptors. Methysergide was administered to rats 10 min prior to DMT and the test was carried out immediately after the injection of DMT for 40 min. Furginele *et al.* [18] found that 15 mg/kg or higher doses of cinanserin, given 10–20 min before hexobarbital, significantly increases hexobarbital-induced sleeping time in mice; pretreatment 30 min with 20 mg/kg of the serotonin antagonist also blocked the EEG effect of 10 mg/kg of 5-hydroxytryptophan.

The adrenergic α-receptor blocker, phentolamine hydrochloride (5 and 10 mg/kg), and the β-receptor blocker, propranolol hydrochloride (10 and 15 mg/kg), were administered to rats 15 min before amphetamine sulfate or saline. The two drugs (phentolamine, 5 mg/kg; propranolol, 15 mg/kg), have been given 30 min before amphetamine to observe the effect on the duration of amphetamine stereotyped behavior [23]. Chlorpromazine hydrochloride (4 mg/kg), a catecholaminergic receptor blocker, and DL-α-methyl-*p*-tryosine methyl ester hydrochloride (AMPT, 250 mg/kg), an inhibitor of the synthesis of catecholamines, in saline were injected 2 and 4 hr respectively prior to amphetamine or saline. This same dose of chlorpromazine has been reported to cause in rats a depression in reinforcement rate and a decrease in skin temperature for 2 hr [51], and 250 mg/kg dose of AMPT reportedly reproduced reduction in rat brain dopamine and norepinephrine about 50 and 70 percent respectively at 4 hr [13].

The dopaminergic receptor blocker, pimozide (1 mg/kg), in 30 percent propylene glycol was given 30 min before amphetamine. The same dose of pimozide has been found to antagonize the locomotor stimulation in mice induced by L-DOPA or d-amphetamine [34]. Since Hill and Horita [24] reported that rabbits, after injection with 0.5 mg/kg of pimozide, exhibited sedation and catalepsy as early as 30 min, in our laboratory we have pretreated rats 30 min with 1 mg/kg of pimozide and found antagonism of the hyperthermia induced by 2,5-dimethoxy-4-methylamphetamine, 2,5-dimethoxy-4-ethylamphetamine, or methylphenidate, as well as the locomotor stimulation induced by N^6, $O^{2'}$-dibutyryl adenosine 3′, 5′-monophosphate (cyclic AMP) (Ho, *et al.*, to be published).

(b) Agonists. The second category of agents were given to the trained animals to test for generalization of the d-amphetamine-induced discriminative responding. These compounds are known to either mimic the action of a particular monoamine at postsynaptic sites or act on the release of the amine. Results are expressed as the percent of total responses on the amphetamine lever.

Cholinomimetic oxotremorine sesquifurmarate (0.1 and 0.25 mg/kg) and the ganglionic stimulant, nicotine sulfate (0.3 mg/kg), both in saline, were injected 15 min prior to the extenction test. Cox and Potkonjak [15] have reported a significant increase of rat brain acetylcholine (ACh) concentration 15 min after the administration of 0.5 mg/kg of oxotremorine and the level returned to control value after 30 min. The oxotremorine-induced tremor increased as the dosage was increased from 0.125 to 1 mg/kg. Morrison and Stephenson [35] employed 0.2 mg/kg of nicotine to establish conditioned stimulus in discriminative learning. Utilizing various tests [8, 19, 33], it has been reported that 0.2 mg/kg of nicotine injected 10 or 15 min before testing greatly facilitated the learning of rats.

The dopamine (DA) receptor stimulant, apomorphine hydrochloride, in dosages ranging from 0.25 to 2 mg/kg in saline, was given to rats 15 min prior to testing. In the study of Maj *et al.* [34], AMPT was used to observe changes in the locomotor stimulation induced by apomorphine (1 and 2 mg/kg, 15 min before test).

Rats also received (a) amantadine hydrochloride (25 and 50 mg/kg), a DA releaser, in saline 45 min before test and (b) the precursor of DA, L-DOPA (50 and 100 mg/kg) in 0.5 percent methylcellulose 30 min before test. The amino acid was given after a 30 min pretreatment using 50 mg/kg of peripheral decarboxylase inhibitor, Ro 4-4602. In addition, a combination of Ro 4-4602, amantadine and L-DOPA was also tested. The effect of amantadine on L-DOPA-induced accumulation of norepinephrine (NE) and DA had been studied in mice [48]. Amantadine (50 and 100 mg/kg) given 105 min before sacrifice, caused only a decrease in the NE accumulation in brain after L-DOPA; while an enhancement of the accumulation of DA was observed after 25 mg/kg of amantadine. L-DOPA (150 mg/kg, given 75 min before sacrifice) in combination with Ro 4-4602 (given 30 min before L-DOPA) produced a significant increase in the rate of avoidance responding in rats, and a 7- to 8-fold increase of brain DA over the control value.

A two-sample rank (Mann-Whitney U) test [20] was used to determine the statistical significance of difference between the percent of amphetamine lever choice response following the test drugs shown in Table 1. The level of significance selected was $p<0.05$ (two tailed test).

RESULTS

The effects of various neurochemical agents on the discriminative response control by amphetamine are shown in Table 1. Table 2 presents the average total responses (mean ± S.E.M.) made on both levels during the 5 and 10 min extension tests. In some cases the rate of response decreased upon administration of neurochemical agents; however, the treatments did not appear to affect the discriminability of amphetamine and saline (see Table 1).

In pretreatment studies, these agents in the appropriate vehicle did not disrupt the saline-lever responses when tested at one or more dose levels in the absence of amphetamine (Table 1). The cholinergic receptor blocker, atropine, failed to block d-amphetamine lever responses. Neither the cholinomimetic oxotremorine ($p<0.02$ different from the amphetamine control, see Table 1) nor the ganglionic stimulant nicotine ($p<0.04$, 5 min; $p<0.02$, 10 min) mimicked the amphetamine responses. The degree of amphetamine discrimination was not affected by the treatment with either of the two serotonin antagonists, methysergide and cinanserin. Neither the alpha nor the beta adrenergic blockers, phentolamine and propranolol respectively, attenuated the percent of amphetamine lever choice in rats. Chlorpromazine, an agent capable of blocking both receptors of noradrenergic and dopaminergic nervous systems, weakened the discriminability of amphetamine by lessening the responses on the amphetamine lever; however, the amphetamine lever responses of the chlorpromazine pretreatment group were not statistically different from the responses of either amphetamine or the saline control groups. Further reduction was seen by the use of pimozide, a dopamine receptor blocker. Animals pretreated with this blocker responsed to the amphetamine lever at only about a 30 percent level during the 10 min intervals ($p<0.01$ different from the amphetamine control).

Attempts to generate the d-amphetamine stimulus condition by the DA receptor stimulant, apomorphine, were unsuccessful. A low dose of 0.25 mg/kg of the drug was apparently too weak to produce an amphetamine-like response ($p<0.02$). Increasing doses to 1 and 2 mg resulted in the inability of some of the animals to press the lever as was observed in 2 out of 5 rats. With 1 mg/kg, the responses of animals at the first 5 min testing period were different from those of either amphetamine control or saline control groups, although the responding animals showed amphetamine lever responses in the 10 min interval.

L-DOPA in the presence of the peripheral decarboxylase inhibitor, Ro 4-4602, produced the amphetamine-like response at a dose of 100 mg/kg ($p<0.04$ and 0.02 different from the saline control at 5 and 10 min respectively) but not at 50 mg/kg. Amantadine alone did not show the same stimulus property as d-amphetamine; animals receiving a dose as high as 50 mg/kg failed to make any response during the first 5 min extinction test and showed saline-like responses which were different from those with d-amphetamine ($p<0.03$) in the second 5 min interval. A combination of amantadine, L-DOPA and Ro 4-4602 caused amphetamine-like responses in rats. The blockade of the synthesis of catecholamines by α-methyltyrosine prevented the discriminative control in rats by d-amphetamine.

DISCUSSION

The rssults of the present study show, with the aid of a number of conventionally used neurochemical agents, that

TABLE 1

EFFECTS OF VARIOUS NEUROCHEMICAL AGENTS ON DISCRIMINATIVE RESPONSE CONTROL BY *d*-AMPHETAMINE

Pretreatment Drug	Test Drug[a]	Percent Amphetamine Lever Choice[b] 5 min	10 min
–	Saline	19.6 ± 4.7	16.1 ± 1.1
–	*d*-Amphetamine	90.5 ± 5.0	88.1 ± 3.2
Atropine (1 mg/kg)	*d*-Amphetamine	96.0 ± 3.1	96.7 ± 2.1
(2 mg/kg)	*d*-Amphetamine	94.6 ± 5.4	92.6 ± 5.3
Atropine (1 mg/kg)	Saline	10.2 ± 2.0	12.0 ± 2.0
(2 mg/kg)	Saline	14.0 ± 5.4	21.6 ± 6.2
–	Oxotremorine		
	(0.25 mg/kg)	23[c]	22[d]
	(0.1 mg/kg)	35.5 ± 3.3*	42.4 ± 4.6*†
–	Nicotine		
	(0.3 mg/kg)	41.0 ± 4.8**	29.7 ± 9.7*
Methysergide (4.5 mg/kg)	*d*-Amphetamine	97.4 ± 0.9	97.8 ± 0.7
(4.5 mg/kg)	Saline	15.0 ± 1.2	13.6 ± 1.7
Cinanserin (15 mg/kg)	*d*-Amphetamine	84.2 ± 4.5	87.2 ± 5.0
(15 mg/kg)	Saline	16.2 ± 6.7	21.5 ± 4.6
Phentolamine (10 mg/kg)	*d*-Amphetamine	93.7 ± 6.4[e]	95.3 ± 4.7[e]
(5 mg/kg)	*d*-Amphetamine	97.4 ± 1.7	97.4 ± 1.7
Phentolamine (10 mg/kg)	Saline	10.2 ± 5.3	9.5 ± 5.5
(5 mg/kg)	Saline	20.0 ± 3.4	20.0 ± 3.4
Propranolol (15 mg/kg)	*d*-Amphetamine	98.6 ± 1.4	98.4 ± 1.0
(10 mg/kg)	*d*-Amphetamine	91.0 ± 3.7	91.0 ± 3.7
Propranolol (15 mg/kg)	Saline	52.4 ± 14.8	51.2 ± 12.6***
(10 mg/kg)	Saline	14.8 ± 6.8	14.8 ± 6.8
Chlorpromazine (4 mg/kg)	*d*-Amphetamine	51.9 ± 7.3	54.9 ± 7.6
(4 mg/kg)	Saline	15.0 ± 5.1	15.0 ± 5.3
Pimozide (1 mg/kg)	*d*-Amphetamine	40.6 ± 17.4*	26.2 ± 11.3***
(1 mg/kg)	Saline	17.6 ± 7.9	19.0 ± 8.9
Propylene Glycol (30%)	*d*-Amphetamine	97.5 ± 1.5	98.5 ± 1.0

the discriminative stimulus properties of d-amphetamine were not mediated by central cholinergic and serotonergic systems. By the intraventricular administration of d-amphetamine, it has previously been domonstrated that the production of discriminative responding is due to the action of d-amphetamine on the CNS [38,39].

Various central effects of d-amphetamine have been shown to be a result of affecting the central catecholaminergic systems in which both NE and DA are involved. The lack of an effect of alpha and beta adrenergic blockers on the amphetamine lever responses in this study failed to establish a role of noradrenergic neurons in the d-amphetamine-induced discriminative control.

Hornykiewicz [27] suggested that DA, and particularly the striatal DA, is involved in the amphetamine syndrome. The involvement of DA in the amphetamine action was first demonstrated with chlorpromazine, which exerts blocking action on both NE and DA receptors [2, 3, 12, 14]. In the present study, chlorpromazine tended to decrease the ability of rats to discriminate d-amphetamine. This stimulus-state associated with d-amphetamine was also lowered by pretreatment with another DA blocker, pimozide. The action of pimozide has been shown to be selective for brain DA receptors [1], although a more recent report indicated an antagonism by pimozide of NE-induced elevation of cyclic AMP content in slices of the rat limbic forebrain [7].

L-DOPA, the precursor of NE and DA, when given to rats at 100 mg/kg dose, increases brain DA while having little effect on the NE level [11]. Brain DA can be further increased more than 3-fold by a combination of L-DOPA and Ro 4-4602 [11]. The latter, when given orally or parenterally to animals, has been shown to markedly inhibit the aromatic amino acid decarboxylase in extracerebral tissues but to exert little effect on the enzyme in brain [5, 6, 36]. As a result Ro 4-4602 facilitates the passage of L-DOPA into the cerebral tissues. Generalization to the d-amphetamine state was demonstrated, as shown in Table 1, using 100 mg/kg of L-DOPA with a pretreatment of 50 mg/kg of Ro 4-4602, and also by adding 50 mg/kg of

TABLE 1 (CONTINUED)

EFFECTS OF VARIOUS NEUROCHEMICAL AGENTS ON DISCRIMINATIVE RESPONSE CONTROL BY *d*-AMPHETAMINE

Pretreatment Drug	Test Drug[a]	Percent Amphetamine Lever Choice[b] 5 min	10 min
–	Apomorphine		
	(2 mg/kg)	32.7 ± 14.2[f]	40.6 ± 14.1[f]
	(1 mg/kg)	57.7 ± 15.0[f]	57.5 ± 13.1[f]‡
	(0.5 mg/kg)	50.8 ± 10.8*	56.2 ± 14.3
	(0.25 mg/kg)	29.0 ± 5.7*	30.2 ± 5.3*
Ro 4-4602 (50 mg/kg)	L-DOPA		
	(50 mg/kg)	57.3 ± 8.7§	52.7 ± 4.4§
(50 mg/kg)	(100 mg/kg)	83.0 ± 11.9††	76.2 ± 4.0‡‡
(50 mg/kg)	Methylcellulose (0.5%)	12.0 ± 2.7	18.4 ± 1.7
Ro 4-4602 (50 mg/kg) + Amantadine (50 mg/kg)	L-DOPA		
	(100 mg/kg)	100[g]	84[g]
Ro 4-4602 (50 mg/kg) + Amantadine (25 mg/kg)	L-DOPA		
	(100 mg/kg)	73.0 ± 9.0‡‡	73.0 ± 13.1‡‡
–	Amantadine		
	(50 mg/kg)	0[h]	51.0 ± 14.3****
	(25 mg/kg)	27.4 ± 13.5	27.4 ± 13.7
α-Methyltyrosine (250 mg/kg)	*d*-Amphetamine	11.5 ± 4.8	16.2 ± 7.3
(250 mg/kg)	Saline	19.0 ± 10.7	16.0 ± 5.9

[a]The dose of *d*-amphetamine sulfate was 0.8 mg/kg, that of saline 1 ml/kg, and all the others were as indicated.

[b]Rats, after being trained for discriminative responding to *d*-amphetamine, were injected intraperitoneally with various doses of the drugs, and the 10 min extinction test was performed at various times as specified in Method. Correct responses were judged by observing responses on the amphetamine lever during 5 and 10 min periods. Each value, unless otherwise specified, represents the mean (± SEM) of 5 determinations in each group of 5 animals. Most of the animals were used more than once but not on the same generalization test. Numbers of animals in a group of 5 made responses during the extinction test: [c]1 on first 5 min; [d]2 on the second 5 min; [e]4 on all 10 min; [f]3 on all 10 min; [g]1 on all 10 min; [h]none on first 5 min.

Significantly different from *d*-amphetamine control group: *$p<0.02$ **$p<0.04$ ***$p<0.01$ ****$p<0.03$

Significantly different from saline control group: †$p<0.01$ ‡$p<0.03$ ††$p<0.04$ ‡‡$p<0.02$

Significantly different from both *d*-amphetamine and control groups: §$p<0.04$

amantadine to the combination. It has been reported that amantadine releases catecholamine from neuronal pools [4, 41, 48], inhibits the uptake of catecholamines into central nerve endings [17] and also increases DA synthesis by increasing the rate of conversion of tyrosine into DOPA [41]. Any of these actions would be anticipated to potentiate the formation of central DA from L-DOPA. In fact, in one animal amantadine at 50 mg/kg accentuated the amphetamine-like cueing effect of L-DOPA and Ro 4-4602 over that produced by L-DOPA and Ro 4-4602 alone. Unfortunately, only the one animal responded to the lever at this dose, and a lower dose of 25 mg/kg did not yield amphetamine-like responses.

The antagonism of d-amphetamine responding by pretreatment with α-methyltyrosine, an inhibitor of catecholamine synthesis [47,52], suggests that the discriminative control by d-amphetamine in rats is produced via the release of newly synthesized DA rather than via a direct action of d-amphetamine on the receptors. Thus, the data from this particular behavior paradigm indicate an indirect central action of amphetamine and are not inconsistent with other reports on the behavioral effects of d-amphetamine [16, 37, 40, 49, 52, 53]. It is difficult to provide an explanation on the failure by apomorphine to exhibit the stimulus property of amphetamine.

In conclusion, the present study has elucidated the role of DA in d-amphetamine induced discriminative responding. The uncertainty of the involvement of NE in this stimulus state was revealed by the failure of both α- and β-adrenergic blockers to affect the amphetamine-like responses. While this manuscript was under revision, Kuhn *et al.* [32] also reported the disruption of d-amphetamine-saline discrimination by AMPT. Although AMPT antagonized discrimination behavior induced by both d-amphetamine and nicotine, the dissimilarity in the stimulus property of the two compounds has been demonstrated [35,44]. Judging from these observations, the above mentioned authors proposed mediation of the amphetamine effect via dopaminergic pathways.

TABLE 2

EFFECTS OF VARIOUS NEUROCHEMICAL AGENTS ON THE RESPONSE RATE

Pretreatment Drug	Test Drug*	Average Total Responses† 5 min	10 min
–	Saline	37.6 ± 6.0	56.8 ± 9.8
–	*d*-Amphetamine	23.6 ± 2.2	43.8 ± 3.1
Atropine (1 mg/kg)	*d*-Amphetamine	25.5 ± 4.8	43.7 ± 6.9
(2 mg/kg)	*d*-Amphetamine	16.8 ± 4.0	29.2 ± 7.2
Atropine (1 mg/kg)	Saline	38.8 ± 4.3	66.0 ± 7.9
(2 mg/kg)	Saline	26.0 ± 2.5	39.2 ± 3.8
–	Oxotremorine		
	(0.25 mg/kg)	26	28
	(0.1 mg/kg)	33.0 ± 6.2	50.8 ± 9.5
–	Nicotine		
	(0.3 mg/kg)	25.5 ± 4.6	43.0 ± 5.1
Methysergide (4.5 mg/kg)	*d*-Amphetamine	32.8 ± 7.3	57.8 ± 12.6
(4.5 mg/kg)	Saline	43.2 ± 7.6	76.2 ± 10.7
Cinanserin (15 mg/kg)	*d*-Amphetamine	26.5 ± 3.7	44.7 ± 6.3
(15 mg/kg)	Saline	27.2 ± 2.9	46.0 ± 5.8
Phentolamine (10 mg/kg)	*d*-Amphetamine	9.0 ± 1.2	16.0 ± 4.0
(5 mg/kg)	*d*-Amphetamine	23.2 ± 6.3	36.2 ± 7.9
Phentolamine (10 mg/kg)	Saline	25.0 ± 5.9	49.7 ± 14.4
(5 mg/kg)	Saline	24.0 ± 2.0	41.8 ± 4.9
Propranolol (15 mg/kg)	*d*-Amphetamine	14.2 ± 5.5	29.7 ± 7.8
(10 mg/kg)	*d*-Amphetamine	28.7 ± 7.0	39.7 ± 9.5
Propranolol (15 mg/kg)	Saline	16.6 ± 2.5	34.4 ± 2.2
(10 mg/kg)	Saline	22.0 ± 10.0	36.8 ± 7.2
Chlorpromazine (4 mg/kg)	*d*-Amphetamine	28.6 ± 0.5	45.6 ± 2.4
(4 mg/kg)	Saline	29.5 ± 4.3	45.8 ± 8.7
Pimozide (1 mg/kg)	*d*-Amphetamine	31.2 ± 5.8	61.2 ± 10.4
(1 mg/kg)	Saline	17.0 ± 2.9	44.4 ± 21.5
Propylene Glycol (30%)	*d*-Amphetamine	29.2 ± 4.0	51.6 ± 8.8
–	Apomorphine		
	(2 mg/kg)	26.3 ± 3.4	41.3 ± 8.1
	(1 mg/kg)	24.3 ± 4.3	46.5 ± 9.1
	(0.5 mg/kg)	36.1 ± 3.4	59.4 ± 4.4
	(0.25 mg/kg)	27.0 ± 2.9	45.0 ± 5.8
Ro 4-4602 (50 mg/kg)	L-DOPA		
	(50 mg/kg)	17.3 ± 4.1	27.0 ± 7.6
(50 mg/kg)	(100 mg/kg)	10.0 ± 4.5	18.5 ± 7.5
(50 mg/kg)	Methylcellulose (0.5%)	24.4 ± 1.4	39.4 ± 2.5
Ro 4-4602 (50 mg/kg) + Amantadine (50 mg/kg)	L-DOPA		
	(100 mg/kg)	9	25
Ro 4-4602 (50 mg/kg) + Amantadine (25 mg/kg)	L-DOPA		
	(100 mg/kg)	8.75± 1.1	16.0 ± 2.3

TABLE 2 (CONTINUED)

EFFECTS OF VARIOUS NEUROCHEMICAL AGENTS ON THE RESPONSE RATE

Pretreatment Drug	Test Drug*	Average Total Responses† 5 min	10 min
–	Amantadine (50 mg/kg)	11.0	19.0 ± 2.1
	Amantadine (25 mg/kg)	72.2 ± 15.1	142.8 ± 34.0
α-Methyltyrosine (250 mg/kg)	*d*-Amphetamine	27.4 ± 6.6	44.2 ± 12.4
(250 mg/kg)	Saline	7.2 ± 1.1	14.0 ± 1.6

*The dose of *d*-amphetamine sulfate was 0.8 mg/kg, that of saline 1 ml/kg, and all the others were as indicated.

†Each value represents the mean (± SEM) of total responses made on both levers during the 5 and 10 min extinction tests. The number of animals which made responses on the levers is shown in footnote b of Table 1.

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