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Blocking effect of α-methyltyrosine on amphetamine based reinforcement

The intravenous injection of amphetamine-like drugs into man causes characteristic euphoric sensations which are commonly regarded as the basis for drug dependence of the amphetamine type (Eddy, Halbach, & others, 1965). Recent clinical studies (Jönsson, Gunne & Anggard, 1969; Jönsson, Anggard & Gunne, 1971) have shown this subjective euphoric action of (±)-amphetamine to be blocked by α-methyltyrosine, an inhibitor of tyrosine hydroxylase which had previously been found to block the behavioural effects of amphetamine in laboratory species (Weissman & Koe, 1965; Hanson, 1966; Randrup & Munkvad, 1966). Although the euphoric effect of amphetamine-like drugs has been equated with their experimental effect of serving as primary positive reinforcers (Wikler, 1971; Renault & Schuster, 1972; Crowley, 1972), it has not been shown experimentally that α-methyltyrosine (α-MT) can block the reinforcing action of such drugs. Modification of the self-administration behaviour of rats for methamphetamine by α-MT treatment has been reported by Pickens, Meisch & Dougherty (1968). While the authors tentatively suggested that behaviour alterations seen were attributable to a reduction by α-MT of the effectiveness of methamphetamine as a reinforcer, no firm conclusion could be drawn from their preliminary investigation concerning the basis for the observed effects.

There are a number of problems in studying an influence of one drug on the reinforcing effect of another by techniques which measure effects simply in terms of increases or decreases from ongoing operant self-administration baselines. To obviate ambiguities that arise in the interpretation of such results, the present experiments utilized a two-phase design new to self-administration studies (Davis & Smith, 1972). In the first phase, effects of a test agent on primary reinforcement are assessed

on acquisition of the operant behaviour rather than on an established behavioural baseline. If an inhibition of acquisition is found, the second phase is conducted. This phase determines the ability of a test agent to affect the development of a Pavlovian based conditioned reinforcer. Such development is reflected in an operant response measure performed later, subsequent to termination of the action of the test drug, allowing recovery from its immediate effects.

Adult male Holtzman albino rats, 350 to 400 g were used. Between sessions they were housed individually in a room separate from the experimental area. Both in the home cage and in the experimental apparatus, food and water were freely available. The rats under ether anaesthesia were implanted with a jugular cannula and connected via an external leather and metal "saddle" to a "leash" consisting of a 30·5 cm length of flexible metal needle tubing and a swivel. These were attached in turn to a pivoting horizontal "arm" above the experimental chamber. Passing through this arm was a plastic tubing connecting to the syringe-driver system which delivered infusions. Programming equipment permitted delivery of a small volume of drug solution either involuntarily on an automatic schedule, or voluntarily as the consequence of an operant response, i.e., the depression of a bar. This bar could be removed or inserted through an aperture in one side of the experimental chamber which consisted of a plexiglass cylinder (diameter 25 cm, height 24 cm) with a wire mesh floor. At least 24 h elapsed after surgery before the animals were used.

In each of the two experiments, 1 h was allowed for rats to adapt to the experimental chambers, following which a 6 h operant level was determined on Day 1. During this time each bar-press resulted in a 0·2 s intravenous infusion of 0·018 ml of 0·9% saline solution coinciding with a 0·2 s buzzer presentation. In Experiment I on Day 2 a 6 h acquisition period was run with all conditions the same as Day 1 except that a 15 μg kg^{-1} dose of (+)-amphetamine sulphate was substituted for saline. These conditions were applied in order to discriminate any "saline responder" or any subject not evidencing reinforcement from this dose of amphetamine. On Day 3, a 6 h extinction period was given in which all conditions of Day 1 were reinstated. On the 4th day, 3 intraperitoneal injections were given at 4 h intervals, each of 75 mg kg^{-1} of L-α-methyl-*p*-tyrosine (α-MT; Regis Chemical Co.) suspended in 0·9% saline, or an equal volume of saline solution. Fifteen min after the 3rd injection, a 6 h reacquisition period began during which the same contingencies as in the 1st acquisition prevailed. This tested the capacity of α-MT to block acquisition of (+)-amphetamine self-administration behaviour.

In Experiment II on the 2nd day, 3 intraperitoneal injections were given at 4 h intervals, each of either 75 mg kg^{-1} α-MT or an equal volume of saline. Fifteen min after the 3rd injection subjects were placed in the experimental chambers with the bars removed, and 50 buzzer-amphetamine sulphate pairings were given without regard to the subjects' behaviour. The amphetamine dose was 15 μg kg^{-1}, and infusion volume and duration were the same as previously. Infusions during the non-contingent pairings were programmed so that during every 6 min either 1, 2, 3, 4 or 5 injections (paired with buzzer) were administered. The variable injection frequency occurred randomly throughout the pairing period which lasted approximately 100 min. Four days after buzzer-amphetamine sulphate pairings, the rats were replaced in the test chamber with operant period (Day 1) conditions reinstated. If the buzzer had acquired reinforcing properties, increases in bar-pressing over the operant level would be expected; therefore, this period tested for the establishment of conditioned reinforcement, and for the blocking of such by α-MT. On the following (7th) day, 6 h period was given in which bar-pressing led to the buzzer plus the same (+)-amphetamine dose as given during pairings. This was to discriminate any rat that would not respond to this dosage of amphetamine sulphate as a primary

Table 1. *Effect of α-methyltyrosine pretreatment (3 intraperitoneal doses of 75 mg kg^{-1}) on reacquisition of an extinguished barpress response for reinforcement consisting of 15 μg kg^{-1} per infusion of (+)-amphetamine sulphate.*

Treatment Group	No. of Rats	Mean bar-press responses (± s.e.) Operant Level	Initial Acquisition	Reacquisition
Saline	8	34 ± 9	81 ± 21	214 ± 44
α-Methyltyrosine	7	45 ± 14	85 ± 21	38 ± 11*

* Significant difference from saline response ($P < 0.001$).

Table 2. *Effect of α-methyltyrosine pretreatment (3 intraperitoneal doses of 75 mg kg^{-1}) before pairings at day 2 of buzzer and non-contingent infusions of (+)-amphetamine sulphate (in a dose of 15 μg kg^{-1}) on the occurrence of conditioned reinforcement measured by bar-pressing for buzzer plus saline infusion at day 6.*

Treatment Group	No. of rats	Mean bar-press responses (± s.e.) Operant level	Conditioned reinforcement
Saline	8	37 ± 6	113 ± 12
α-Methyltyrosine	8	43 ± 9	39 ± 9*

* Significant difference from saline response ($P < 0.005$).

reinforcer, since such subjects could not be expected to develop conditioned reinforcement.

The data of Experiment I (Table 1) show that operant levels for the saline and α-MT groups did not differ significantly according to the Mann-Whitney U Test (Siegel, 1956). Initial acquisition of amphetamine self-administration behaviour on Day 2 was almost identical for the 2 groups. However, after extinction, reacquisition data show that the response level of the α-MT group did not differ from operant level, and was significantly less than the saline group ($P < 0.001$). α-MT clearly blocked an action of (+)-amphetamine essential to reacquisition of the bar-press behaviour. In Experiment II (Table 2) the operant levels again were similar for both groups, whereas the saline group responded 3 times as often as the α-MT group ($P < 0.005$) in the test for conditioned reinforcement. This indicates a blocking of the primary reinforcing action of amphetamine by α-MT during the amphetamine sulphate-buzzer pairings. A Wilcoxon Matched Pairs Test (Siegel, 1956) showed that the difference between operant level and performance in the test of conditioned reinforcement was not significant ($P > 0.05$) for the α-MT group, while the difference was significant for the saline group ($P < 0.01$). Thus, data for the α-MT group give no indication of motor depression, but indicate effective reinforcer blocking, whereas strong conditioned reinforcement was observed in the saline group.

The inconclusiveness of the earlier study of α-MT on methylamphetamine self-administration (Pickens & others, 1968) stems from methodological considerations alluded to above plus a particular alternative explanation to that of a specific action on reinforcement. Namely, the α-MT might have acted by antagonizing certain side effects or toxicity of self-administered methylamphetamine, especially with the much higher unit dosage used (500 μg kg^{-1}), which may tend to retard the self-administration behaviour. This antagonism would yield an initially elevated rate of methylamphetamine intake. Proportionately to such occurrence, there should follow a

period of response inhibition because of the "extra" intake of the stimulant. An effect of this sort has been demonstrated by Pickens & others (1968) with intraperitoneal doses presented during self-administration of methylamphetamine. Because of the different design of the present study, this possible explanation cannot account for the effectiveness of α-MT in preventing both reacquisition of self-administration behaviour and the development of conditioned reinforcement. In both instances bar-pressing much above the operant level occurred in saline control rats because of the direct or indirect functioning of the primary reinforcing property of (+)-amphetamine. In neither case was such bar-pressing behaviour acquired in the α-MT rats.

The antagonism of the positive reinforcing property of (+)-amphetamine that we have found is in accordance with the view that drugs of this type act through a neurochemical mechanism which is functionally dependent on a critical pool of brain catecholamines (for recent review see, e.g., van Rossum, 1970; Moore & Dominic, 1971). The results may represent in some degree an equivalent of the clinical findings of Jönsson, Anggard & Gunne (1971). Both the laboratory and clinical data suggest that α-MT or similar drugs have potential value in treatment of drug abuse of the amphetamine type. If pharmacological block of the α-MT type were maintained, and if then stimulant self-administration occurred in the usual circumstances of drug use, two benefits should result: (1) drug-taking behaviour maintained by primary reinforcement should be extinguished, and (2) conditioned (secondary) reinforcers, i.e., environmental stimuli associated with drug abuse, also should lose their effectiveness because of non-reinforcement.

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