EUROPEAN JOURNAL OF PHARMACOLOGY 19 (1972) 25–34. NORTH-HOLLAND PUBLISHING COMPANY

EFFECTS OF 5-METHOXY-N,N-DIMETHYLTRYPTAMINE ON CENTRAL MONOAMINE NEURONS

Kjell FUXE, Bo HOLMSTEDT and Gösta JONSSON

Department of Histology, Karolinska Institutet, 104 01 Stockholm, and Department of Toxicology, Swedish Medical Research Council, Karolinska Institutet, 104 01 Stockholm, Sweden

Received 7 October 1971 Accepted 21 February 1972

K. FUXE, B. HOLMSTEDT and G. JONSSON, *Effects of 5-methoxy-N,N-dimethyltryptamine on central monoamine neurons*, European J. Pharmacol. 19 (1972) :25–34.

Using α-propyldopacetamide (H 22/54), a tryptophan hydroxylase inhibitor, and α-methyltyrosine methylester (H 44/68), a tyrosine hydroxylase inhibitor, the effects of 5-methoxy-N,N-dimethyltryptamine (5-MeO-DMT) on amine turnover in central 5-HT, NA and DA nerve terminals of the rat were studied using histochemical fluorescence methods for the demonstration of CA and 5-HT. It was found that 5-MeO-DMT in single or repeated doses of 5 mg/kg decreased the H 22/54-induced disappearance of 5-HT fluorescence in 5-HT nerve terminals and increased the H 44/68 and H 22/54-induced disappearance of NA fluorescence in NA nerve terminals, suggesting that 5-MeO-DMT decreased 5-HT turnover and increased NA turnover. In the functional experiments, 5-MeO-DMT markedly increased the extensor hindlimb reflex of the spinal rat independently of presynaptic 5-HT stores, a reflex which is highly dependent on 5-HT receptor activity. These results suggest that 5-MeO-DMT can directly stimulate central 5-HT receptors. It is proposed that the decrease in 5-HT turnover observed is secondary to the 5-HT receptor stimulation induced by 5-MeO-DMT, which elicits a negative feed-back resulting in a reduction of the activity of the 5-HT neurons. In view of previous studies on d-LSD, dimethyltryptamine and psilocybin (Andén et al., 1968, 1971) and the present results, a capacity to stimulate 5-HT receptors seems to be a common property of hallucinogens of the indolealkylamine type.

5-Hydroxytryptamine
Catecholamines
Extensor reflex
5-Methoxy N,N-dimethyltryptamine
Histochemical amine analysis
Amine turnover

1. INTRODUCTION

Gessner and Page (1962) were the first to describe the potent behavioural effects of 5-methoxydimethyltryptamine (5-MeO-DMT) in animals. The ability of 5-MeO-DMT to produce sham rage and tremor was demonstrated by Benington et al. (1965) and Ahlborg et al. (1968). These studies suggest potent actions of 5-MeO-DMT on the central nervous system and in 1964 Holmstedt et al. had shown that 5-MeO-DMT was the chief component of several psychoactive South-American snuffs. Further studies by Holmstedt's group (Agurell et al., 1968, 1969a,b) revealed this drug to be a common constituent of South American hallucinogenic drugs and plants. Pharmacological studies on this drug have mainly been performed by Gessner (1970).

In previous papers (Andén et al., 1968, 1971), hallucinogenic drugs of the indolealkylamine type such as d-lysergic acid diethylamide (LSD), psilocybin and dimethyltryptamine (DMT) were shown to decrease central 5-HT turnover, and this action appeared to be initiated by the 5-hydroxytryptamine (5-HT) receptor stimulation caused by these hallucinogens. There seems to be a certain correlation between 5-HT receptor stimulating activity and hallucinogenic potency. The present investigation was therefore prir arily undertaken to see if 5-MeO-DMT also interfered with 5-hydroxytryptaminergic mechanisms in a similar way.

2. MATERIALS AND METHODS

Male Sprague–Dawley rats (b.wt. 150–170 g) were used. The rectal temperature of the rats was frequently monitored and was found to vary between 36.5 and 37.5°C after injection of 5-MeO-DMT. The drug was synthesized at the Department of Toxicology, Karolinska Institutet and was given i.p. to the rats in saline as the hydrochloride. The doses refer to the base.

2.1. *Amine turnover*

The 5-HT turnover was evaluated with the help of a tryptophan-hydroxylase inhibitor, α-propyldopacetamide (H 22/54; Carlsson et al., 1963; 500 mg/kg, i.p., 3 hr before killing). The rate of decline of the 5-HT stores gives an indication of the 5-HT turnover (see Andén et al., 1969); the doses and number of rats studied are given in table 1. In the majority of cases several injections of 5-MeO-DMT were given, since the effects of the drug on the extensor hindlimb reflex (see below) and on behaviour were short-lasting.

The studies on catecholamine (CA) turnover were performed with the help of a tyrosine-hydroxylase inhibitor, α-methyltyrosine methylester (H 44/68, i.p., 250 mg/kg, 2–4 hr before killing) or H 22/54 (500 mg/kg, i.p. 2 hr before killing). For references, see Udenfriend (1966) and Andén et al. (1966a). The rate of decline of the CA stores gives an indication of CA turnover (see Andén et al., 1969). At the time intervals used in the present study, the results mainly reflect changes in turnover in the large bulk pool of amines; for doses and numbers of rats, see tables 3 and 4.

The rats were killed by rapid decapitation under slight chloroform anaesthesia. The telencephalon, diencephalon, mesencephalon and rhombencephalon were rapidly dissected out and taken to histochemical fluorescence analysis of CA and 5-HT according to Falck and Hillarp (Falck et al., 1962; Hillarp et al., 1966; Corrodi and Jonsson, 1967). The procedure for demonstration of 5-HT and CA was performed as modified by Fuxe and Jonsson (1967) in order to ensure as strong yield as possible of the fluorescent products. For details on the preparation of the paraformaldehyde, see Hamberger et al., 1965 and Hamberger, 1967. 5-MeO-DMT itself does not react with formaldehyde gas to form a fluorescent compound since it is a tertiary amine (Corrodi and Jonsson, 1965). The 5-HT nerve terminals of the nucleus suprachiasmaticus were studied, since this is a well-defined nucleus densely innervated by the 5-HT nerve terminals (Fuxe, 1965). The NA nerve terminals were evaluated mainly in the hypothalamus and the vagal area, areas densely innervated by NA nerve terminals (Fuxe, 1965). The DA nerve terminals were evaluated in the neostriatum. Furthermore, the NA nerve terminals of the cortical areas (neocortex, hippocampal formation, cerebellar cortex) were studied by the smear technique, as described by Olson and Ungerstedt (1970). This procedure is very simple and involves taking small pieces of the cortical areas, smearing them on objective slides followed by drying over phosphorus pentoxide after which the slides were exposed to formaldehyde gas.

A semiquantitative estimation of fluorescence intensity was made on coded slides. It is known that a change in fluorescence intensity reflects a change in amine concentration (Olson et al., 1968; Jonsson, 1969; Lidbrink and Jonsson, 1971). The problems of semiquantitation in amine histochemistry has been thoroughly discussed by Jonsson (1971; see also Fuxe et al., 1970b). The estimation of intensity obtained from one area of an animal represents the mean value of 4–6 observations. Although it is theoretically not correct to calculate mean values from semiquantitative data, the means, in addition to the primary observations, are presented to simplify reading of the tables (see Lidbrink and Jonsson, 1971).

In one set of experiments, 5-HT turnover was analysed by performing chemical analytical determination of 5-HT in whole brain according to the method of Bertler (1961). For details see table 2. In a control experiment 5-MeO-DMT was not found to interfere with the determination of 5-HT during the experimental conditions used as determined in reserpine-pretreated animals (10 mg/kg, i.p., 24 hr before killing).

2.2. *5-HT accumulation following monoamine oxidase inhibition*

The effects of 5-MeO-DMT were evaluated by studying its effects on nialamide (300 mg/kg, i.p.)-induced 5-HT accumulation in central 5-HT neurons after pretreatment with reserpine (10 mg/kg, i.p., 24 hr before killing), since the 5-HT accumulation is

linear up to 4 hr after injections (Meek and Fuxe, 1971). The rats were usually killed 4 hr after nialamide treatment. 5-MeO-DMT was given i.p. in several doses starting 3.5 hr before killing or in a single dose 45 min before killing. The dose of 5-MeO-DMT was 5 mg/kg and 4–5 rats were used in each group.

2.3. *Experiments with H 75/12 (α-ethyl-4-methyl-metatyramine)*

The effects of 5-MeO-DMT on H 75/12-induced depletion of 5-HT were studied (see Carlsson et al., 1969) to reveal possible effects of 5-MeO-DMT on 5-HT uptake. 5-MeO-DMT, 5 mg/kg, was given i.p. 5 min before and 15 min after each H 75/12 injection (two doses of 25 mg/kg, i.p., with a 2-hr interval). Reduction of 5-HT depletion indicates a drug-induced blockade of the 5-HT membrane pump. The animals were taken to histochemical analysis 2 hr after the last H 75/12 injection as described above; 4–6 animals were used in each group.

2.4. *MAO activity*

The effects of repeated doses (5) of 5-MeO-DMT, 5 mg/kg, on MAO (monoamine oxidase) in homogenates of the hypothalamus were studied by the method of Wurtman and Axelrod (1963), which determines MAO activity from the enzymatic conversion of tryptamine-2-^{14}C to indole acetic acid-^{14}C (10.9 mCi/mmole; New England Nucl. Corp.). The drug was given in the same way and at the same time intervals as in the turnover experiments with H 22/54.

Studies on intraneuronal MAO were performed on rats pretreated with reserpine (10 mg/kg, i.p., 24 hr before killing). MAO inhibitors, such as pargyline and nialamide, increase accumulation of intraneuronal 5-HT. Effects of 5-MeO-DMT, 5 mg/kg, in a dose that caused retardation of H 22/54 induced 5-HT depletion (see results) were determined; MAO inhibitors also cause dose-dependent retardation of 5-HT depletion induced in this way, see Andén et al. (1971); 4–6 animals were used in each group.

2.5. *Functional experiments*

The extensor hindlimb reflex of spinalized rats is highly dependent on 5-HT receptor activity (Andén et al., 1964; Andén. 1968; Meek et al., 1970). Thus, 5-hydroxytryptophan (5-HTP) and tryptophan enhance this reflex and mimic the effects endogenously released 5-HT but not NA. Furthermore, peripheral and circulatory effects obtained by injections of 5-HTP and indolealkylamines are unlikely to interfere with the reflex activity (see Andén et al., 1971).

The rats were acutely spinalized 2–3 hr before injection of the 5-MeO-DMT. The animals were pretreated with reserpine (10 mg/kg, i.p., 24 hr before testing) and H 22/54 (500 mg/kg, i.p., 2 hr before testing) to deplete all known 5-HT stores in the CNS and demonstrate whether the effects of 5-MeO-DMT were dependent on presynaptic 5-HT stores or not. The 5-MeO-DMT was usually injected immediately before testing. In some experiments, NA receptors were blocked by pretreatment with phenoxybenzamine (20 mg/kg, i.p., 1 hr before testing). The extensor hindlimb reflex was induced by pinching the base of the tail. A semiquantitative estimation of the strength of the extensor reflex was made on coded animals; for doses and number of rats, see table 5.

3. RESULTS

3.1. *Turnover studies*

3.1.1. H 22/54 experiments

5-MeO-DMT caused a dose-dependent reduction of H 22/54-induced disappearance of fluorescence from the 5-HT nerve terminals of the nucleus suprachiasmaticus (figs. 1 and 2; table 1); these results were confirmed by chemical determination of 5-HT in whole brain (table 2). H 22/54-induced disappearance of fluorescence in the NA nerve terminals of the hypothalamus and the cortex cerebri (table 3) was increased.

3.1.2. H 44/68 experiments

After repeated doses or a single dose of 5-MeO-DMT, there was a clearcut increase in the H 44/68-induced fluorescence disappearance from the NA nerve terminals of the hypothalamus, the lower brain stem and the cortex cerebri. The results in the hypothalamus are illustrated in table 3 and were similar to those in other parts of the brain. In contrast, the decrease in fluorescence intensity obtained in the DA nerve terminals of the neostriatum after H 44/68 treatment was not significantly influenced by repeated doses of 5-MeO-DMT (table 4).

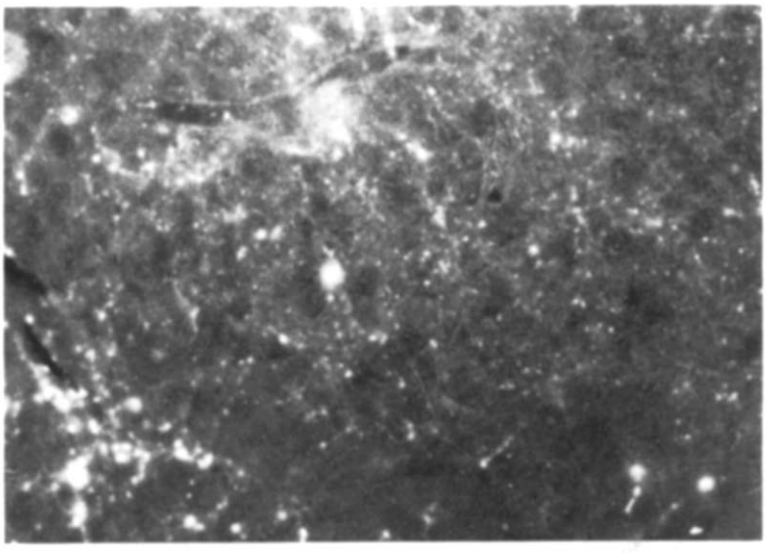

Fig. 1. Nucleus suprachiasmaticus of a rat after treatment with H22/54 (500 mg/kg, 2 hr before killing). Only weakly fluorescent 5-HT nerve terminals are observed (1+ in table 1). The strongly fluorescent large varicosities in the lower left part of the picture represent NA varicosities. × 260.

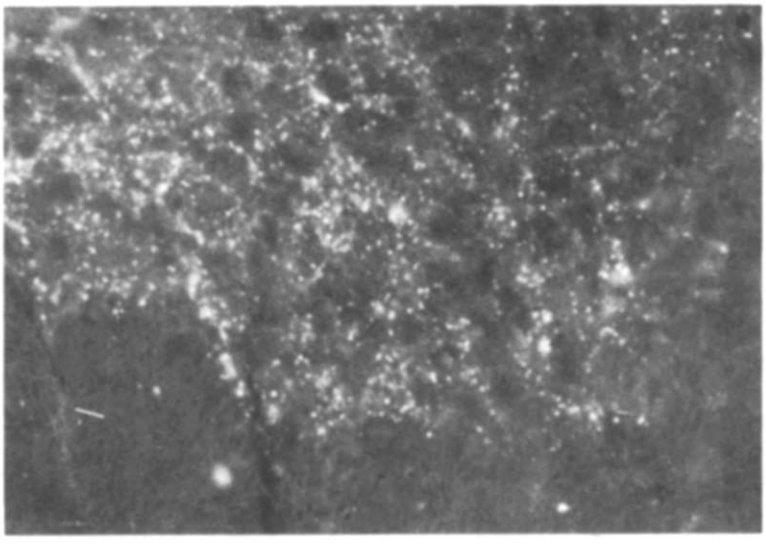

Fig. 2. Nucleus suprachiasmaticus of a rat after combined treatment with 5-MeO-DMT (5 mg/kg, 15 min before the inhibitor) and H22/54 (as described above). Moderately fluorescent 5-HT nerve terminals are observed as fine fluorescent dots due to retardation of 5-HT depletion by 5-MeO-DMT (2+ in table 1). × 260.

Table 1
Effect of 5-MeO-DMT on the H 22/54-induced fluorescence disappearance from the 5-HT nerve terminals of the nucleus suprachiasmaticus.
5-MeO-DMT was given i.p. 15 min before H 22/54 injection (500 mg/kg, i.p. 3 hr before killing). The other 4 doses were given at 30 min intervals starting 30 min following the H 22/54 injection. When only 2 doses were given, the second dose was administered 1 hr after the H 22/54 injection. A semiquantitative estimation of fluorescence intensity was made on coded slides. 3+ = strong intensity, 2+ = moderate intensity, 1+ = weak intensity. When it could not be decided if a fluorescence belonged to one or the other grade, half a plus was added to the lower grade. Number of animals within parentheses. The value for each animal represents the mean value of 4–6 estimations.

Treatment	Fluorescence intensity		Effect on H 22/54-induced fluorescence disappearance
	Means	Range	
No drug treatment	3+	3+(8)	
5-MeO-DMT (5×5 mg/kg)	3+	3+(6)	
5-MeO-DMT (5 mg/kg)	3+	3+(3)	
H 22/54	1+	0.5+(2) 1+(4) 1.5+(2)	
5-MeO-DMT (5×5 mg/kg) + H 22/54	2+	1.5+(2) 2+(3) 2.5+(2)	Moderate retardation
5-MeO-DMT (2×5 mg/kg) + H 22/54	1.6	1+(2) 1.5+(4) 2+(3)	Weak retardation
5-MeO-DMT (5 mg/kg) + H 22/54	1.4	1+(5) 1.5+(5) 2+(2)	Weak reterdation

Table 2
The effects of 5-MeO-DMT on the H 22/54-induced disappearance of 5-HT in whole brain.
5-MeO-DMT (3 × 5 mg/kg, i.p., 1 hr interval) was given i.p. and the first dose 15 min before H22/54 (500 mg/kg, 3 hr before killing). n = number of animals. Statistical evaluation by Student's t-test.

Treatment	n	5-HT (ng/g)	5-HT (%) *
No drug treatment	4	374 ± 13	100
5-MeO-DMT	4	357 ± 24	95
H 22/54	4	214 ± 6 [a]	57
5-MeO-DMT + H 22/54	4	272 ± 24 [b]	73

* Expressed as % of untreated control
[a,b] Difference between a and b: $p < 0.05$.

3.2. *5-HT accumulation following MAO inhibition*

5-MeO-DMT in single or repeated doses (3) of 5 mg/kg given shortly before killing or in several doses starting 3 hr earlier did not change the intraneuronal fluorescence in the cell bodies or in the nerve terminals.

3.3. *Experiments with H 75/12*

5-MeO-DMT did not block H 75/12-induced disappearance of fluorescence from the suprachiasmatic 5-HT nerve terminals.

3.4. *MAO activity*

Repeated doses of 5-MeO-DMT (5 × 5 mg/kg; 30-min intervals), as administered in the H 22/54 experiments, did not inhibit MAO activity in homogenates of the hypothalamus. The MAO activity of the treated group was 114 ± 10 (n = 8) per cent of untreated controls (263 ± 13 c.p.m/mg; n=8). Furthermore, in reserpine-pretreated animals (10 mg/kg, i.p., 24 hr before sacrifice), single or repeated doses of 5-MeO-DMT (5 mg/kg, i.p., first dose injected 2 hr before sacrifice) did not increase fluorescence in the 5-HT cell bodies as did low doses of pargyline (25 mg/kg, i.p., 2 hr before sacrifice) which retarded H 22/54-induced 5-HT depletion as effectively as 5-MeO-DMT (see Andén et al., 1971).

3.5. *Functional experiments*

5-MeO-DMT, 5 mg/kg, caused a marked and short-lasting increase (15–30 min) in the extensor hindlimb reflex activity which was not blocked by previous treatment with reserpine-H 22/54 or by blockade of the α-adrenergic receptors with phenoxybenzamine (table 5). In addition to the increase in the extensor reflex, clear athetoid movements of the hindlimb were observed. At a dose of 1 mg/kg, a clear increase in the extensor lindlimb reflex was observed. A dose of 0.5 mg/kg was hardly effective. The increase in the

Table 3

The effect of 5-MeO-DMT on the H22-54-induced fluorescence disappearance from the NA nerve terminals of the cortex cerebri. 5-MeO-DMT was injected i.p. 15 min before the H 22/54 injection (500 mg/kg, i.p., 2 hr before killing). The other three doses were given at 0.5 hr intervals starting 30 min after the H 22/54 injection. For further details, see text to table 1.

Treatment	Fluorescence intensity		Effect on H 22/54-induced fluorescence disappearance
	Means	Range	
No drug treatment	3+	3+(8)	
5-MeO-DMT (4×5 mg/kg)	3+	3+(4)	
5-MeO-DMT (5 mg/kg)	3+	3+(3)	
H 22/54	1.9+	1.5+(2) 2+(6)	
5-MeO-DMT (4×5 mg/kg) + H 22/54	1.2+	1+(6) 1.5+(3)	Acceleration
5-MeO-DMT (5 mg/kg) + H 22/54	1.2+	1+(5) 1.5+(3)	Acceleration

Table 4

The effect of 5-MeO-DMT on the H 44/68-induced fluorescence disappearance from the NA nerve terminals of the hypothalamus and the DA nerve terminals of the neostriatum.

5-MeO-DMT was given i.p., 15 min before the H44/68 injection (250 mg/kg, i.p., 4 hr before killing). The other three doses were given at 1-hr intervals starting 1 hr following the H44/68 injection. 0.5+ = very weak fluorescence intensity. For further details, see text to table 1 and text in paper.

Treatment	Fluorescence intensity				Effect on H 44/68-induced fluorescence disappearance
	NA		DA		
	Means	Range	Means	Range	
No drug treatment	3+	3+(8)	3+	3+(8)	
5-MeO-DMT (4×4 mg/kg)	3+	3+(6)	3+	3+(6)	
5-MeO-DMT (5 mg/kg)	3+	3+(4)	3+	3+(4)	
H 44/68	1.8+	1+(2) 2+(8)	1.2+	1+(8) 2+(2)	
5-MeO-DMT (4×5 mg/kg) + H 44/68	1.2+	1+(6) 1.5+(3)	1+	0.5+(2) 1+(4)	Acceleration of NA disappearance
5-MeO-DMT (5 mg/kg) + H 44/68	1.4+	1+(4) 2+(3)	1+	1+(4)	Acceleration of NA disappearance

extensor reflex could be maintained by giving repeated doses of the drug.

4. DISCUSSION

5-MeO-DMT caused a dose-dependent decrease in the depletion of 5-HT produced by H 22/54 treatment, observed both histochemically and biochemically. These findings suggest that 5-MeO-DMT decreases 5-HT turnover. The possibility that 5-MeO-DMT could cause an early recovery of 5-HT levels by interfering with amine synthesis inhibition is highly unlikely, since 5-MeO-DMT simultaneously increased H 22/54-induced NA depletion. It is also unlikely that 5-MeO-DMT or its metabolites, interfered with fluorophor formation, since e.g. the drug alone or in combination with reserpine or H 75/12 did not change fluorescence intensity; in a recent metabolic study in rats no metabolite capable of forming a fluorophor was found (Agurell et al., 1969b). Nor is it likely that the retardation induced by 5-MeO-DMT was due to MAO inhibition, since no inhibition of MAO in homogenates of the hypothalamus was observed and the NA depletion induced by H 22/54 was increased. Furthermore, no increases in 5-HT fluorescence were present. A blockade of the 5-HT membrane pump is also known to retard 5-HT turnover

Table 5
Effect of 5-MeO-DMT on the extensor hindlimb reflex of spinal rats.
The spinal cord transection was made in the midthoracic region 2–3 hr before testing. Reserpine (10 mg/kg, i.p.) was given 24 hr before testing and H22/54 (500 mg/kg, i.p.) 2 hr before testing. 5-MeO-DMT was given immediately before testing. Phenoxybenzamine was given 1 hr before testing. The strength of the extensor reflex was semiquantitatively estimated on coded animals. 4+ = very strong; 3+ = strong; 2+ = moderate; 1+ = weak; 0.5+ = very weak. Number of rats within parentheses.

Pretreatment	Treatment	Strength of the extensor reflex	Effect on the reflex by 5-MeO-DMT
–		1+(5)	
–	5-MeO-DMT (5 mg/kg)	4+(6)	Marked increase
Reserpine + H 22/54		0.5+(4)	
Reserpine + H 22/54	5-MeO-DMT (5 mg/kg)	4+(6)	Marked increase
Reserpine + H 22/54	5-MeO-DMT (1 mg/kg)	2+(5)	Increase
Reserpine + H 22/54	5-MeO-DMT (0.5 mg/kg)	1+(4)	
Phenoxybenzamine (20 mg/kg)		1+(4)	
Phenoxybenzamine (20 mg/kg)	5-MeO-DMT (5 mg/kg)	4+(4)	Marked increase
Phenoxybenzamine (20 mg/kg)	5-MeO-DMT (1 mg/kg)	2+(4)	Increase
Reserpine + H 22/54 + phenoxybenzamine (20 mg/kg)		0.5+(4)	
Reserpine + H 22/54 + phenoxybenzamine (20 mg/kg)	5-MeO-DMT (5 mg/kg)	4+(4)	Marked increase

(Corrodi and Fuxe, 1968, 1969). However, such an action can be excluded in this study, since 5-MeO-DMT was shown not to inhibit the 5-HT membrane pump as evidenced by its inability to block H 75/12-induced 5-HT depletion. Blockade by 5-MeO-DMT of the release of 5-HT induced by nervous impulse, which cannot be excluded from the present experiments, would result in a decrease in 5-HT turnover. However, such an action has not been demonstrated, and a reduced 5-HT release by the potent psychotogen, d-LSD has only been demonstrated with relatively high concentration in vitro (Chase et al., 1967; Katz and Kopin, 1969; Farnebo and Hamberger, 1971); after dimethyltryptamine (DMT) 5-HT release is increased (Meek and Fuxe, 1971). Decreased nervous impulse flow in 5-HT neurons is a possible explanation for the observed decrease in 5-HT turnover, since amine turnover is highly dependent on nervous impulse flow (Andén et al., 1966b) and since Aghajanian et a. (1970) have found that the hallucinogen, DMT inhibits electrical activity in the mesencephalic 5-HT raphe cell bodies.

Studies on the effects of 5-MeO-DMT on the extensor hindlimb reflex, which can be used as a gross measure of changes in 5-HT receptor activity (see Method section), indicated that 5-MeO-DMT directly stimulates 5-HT receptor sites. Thus, a marked short-lasting increase in the extensor hindlimb reflex was obtained independent of the presence of presynaptic 5-HT stores. Also the effects on gross behaviour might be due to a 5-HT-like action, since abduction and extension of the hindlimbs were observed together with tremor and head movements, just as in the nialamide-5-HTP syndrome (for recent and earlier references, see Mantegazzini, 1966 and Grahame-Smith, 1971).

It is possible that the 5-HT receptor stimulation induced by 5-MeO-DMT was related to the decreased 5-HT turnover. Thus, 5-HT receptor stimulation caused by 5-MeO-DMT could elicit a feed-back via a chain of neurons ending on the 5-HT cell bodies, in this way inhibiting activity of the 5-HT neurons with a resulting decrease in 5-HT turnover. Part of the feedback could be a local trans-synaptic feedback (cf. Farnebo and Hamberger, 1971), especially in view of recent findings showing that lysergic acid diethylamide can reduce the increase in 5-hydroxy-indolacetic acid levels found upon electrical stimulation of the central 5-HT neurons (Randic and Padjen, 1971).

As with d-LSD, psilocybin and DMT (Andén et al., 1968, 1971; Sugrue, 1969; Leonard and Tonge, 1969), 5-MeO-DMT also increased H 22/54- and

H 44/68-induced NA depletion in various parts of the brain, suggesting an increase in central NA turnover following treatment with 5-MeO-DMT. An increased synthesis of labelled CA from ^{3}H-tyrosine has also been demonstrated following d-LSD treatment (Persson, 1970). NA receptor blockade is known to increase NA turnover (Andén et al., 1967) but is probably not responsible for the present effects, since sedation was not obtained after 5-MeO-DMT or other indolealkylamine hallucinogens; sedation occurs after treatment with known blockers of NA receptors (see book edited by Bobon and Janssen, 1970). The NA neurons are probably part of an arousal system (Fuxe et al., 1970a; Fuxe and Lidbrink, 1971). Activation of NA neurons might be secondary to activation of 5-HT receptors brought about by an interaction between central 5-HT and NA neurons. However, the NA neuronal activation might be a stress reaction in response e.g. to the marked activation of the sympathetic nervous system caused by the hallucinogens (see Cerletti et al., 1968), or to failure of habituation processes (Sheard and Aghajanian, 1968); NA neurons are activated in various types of stress (see e.g. Corrodi et al., 1968; Thierry et al., 1968). Since in sham rage produced by brain stem transection it was possible to relate attack behaviour to NA release in the brain stem (Reis and Fuxe, 1968, 1969), the ability of 5-MeO-DMT to produce sham rage is possibly related to activation of central NA neurons, particularly those in the hypothalamus. Furthermore, EEG arousal found after 5-MeO-DMT could be partly related to activation of the cortical NA neurons, which are probably involved in maintaining tonic cortical arousal (see Jones et al., 1969; Fuxe et al., 1970a; Corrodi et al., 1971).

Previous studies on hallucinogens of the indolealkylamine type have demonstrated decreases of 5-HT turnover (Rosecrans et al., 1967; Anden et al., 1968; Diaz et al., 1968; Lin et al., 1969; Schubert et al., 1970; Freedman et al., 1970; Anden et al., 1971) correlated to electrophysiological findings on the raphe nuclei (Aghajanian et al., 1968, 1970). Since d-LSD and DMT (Aghajanian et al., 1968, 1970) inhibit activity in raphe areas containing 5-HT cell bodies and since d-LSD, psilocybin and DMT (Anden et al., 1968, 1971) probably cause 5-HT receptor stimulation, these drugs might decrease 5-HT turnover via a neuronal feed-back to the 5-HT cell bodies induced by the 5-HT receptor stimulation (Anden et al., 1968, 1971: Aghajanian et al., 1968, 1970). These hallucinogens are probably also capable of stimulating central 5-HT receptors in cat and in man, since d-LSD, psilocybin and DMT have a strong ability to activate the knee-jerk in these species (Isbell 1959; Cerletti et al., 1968). In view of the fact that non-hallucinogenic compounds related to indolalkylamines, such as BOL (2-Br-LSD) and methysergide, do not decrease 5-HT turnover or increase 5-HT receptor stimulation (Anden et al., 1968, 1971), and that BOL is relatively ineffective in decreasing electrical activity in the raphe nuclei (Aghajanian et al., 1970) it seems possible that a capacity to stimulate 5-HT receptors is a common property of hallucinogens of the indolealkylamine type. The present data are in favour of this view and give it further support.

ACKNOWLEDGEMENTS

This work was supported by Grants from the Swedish Medical Research Council, No. B-72-40X-2457-05 (K.F.) and No. B71-40X-2629-03, B71-14X-1021-06 (B.H.), No. B72-14X-2295-05A the National Institutes of Health, GM 13 978, the National Institutes of Mental Health, MH 12007, the Wallenberg Foundation and Riksbankens Jubileumsfond, No. 68/53.

REFERENCES

Aghajanian, G.K., W.E. Foote and M.H. Sheard, 1968, Lysergic acid diethylamide: sensitive neuronal units in the midbrain raphe, Science 161, 706.

Aghajanian, G.K., W.E. Foote and M.H. Sheard, 1970, Action of psychotogenic drugs on single midbrain raphe neurons, J. Pharmacol. Exptl. Therap. 171, 178.

Agurell, S., B. Holmstedt and J.-E. Lindgren, 1968, Alcaloid content of *Banisteryopsis rusbyana*, Amer. J. Pharm. 140, 148.

Agurell, S., B. Holmstedt, J.-E. Lindgren and R.E. Schultes, 1969a, Alcaloids in certain species of *virola* and other South American plants of ethnopharmacologic interest, Acta Chem. Scand. 23, 903.

Agurell, S., B. Holmstedt and J.-E. Lindgren, 1969b, Metabolism of 5-methoxy-N,N-dimethyltryptamine-^{14}C in the rat, Biochem. Pharmacol. 18, 2771.

Ahlborg, U., B. Holmstedt and J.-E. Lindgren, 1968, Fate and metabolism of some hallucinogenic indolealkylamines, Advan, Pharmacol. 6, 213.

Andén, N.-E., 1968, Discussion of serotonin and dopamine in the extra-pyramidal system, Advan. Pharmacol. 6A, 347.

Andén, N.-E., M.G.M. Jukes and A. Lundberg, 1964, Spinal reflexes and monoamine liberation, Nature 202, 1222.

Andén, N.-E., H. Corrodi, A. Dahlström, K. Fuxe and T. Hökfelt, 1966a, Effects of tyrosine hydroxylase inhibition on the amine levels of central monoamine neurons, Life Sci. 5, 561.

Andén, N.-E., K. Fuxe and T. Hökfelt, 1966b, The importance of the nervous impulse flow for the depletion of the monoamines from central neurones by some drugs, J. Pharm. Pharmacol. 18, 630.

Andén, N.-E., H. Corrodi, K. Fuxe and T. Hökfelt, 1967, Increased impulse flow in bulbospinal noradrenaline neurons produced by catecholamine receptor blocking agents, European J. Pharmacol. 2, 59.

Andén, N.-E., H. Corrodi, K. Fuxe and T. Hökfelt, 1968, Evidence for a central 5-hydroxytryptamine receptor stimulation by lysergic acid diethylamide, Brit. J. Pharmacol. 34, 1.

Andén, N.-E., H. Corrodi and K. Fuxe, 1969, Turnover studies using synthesis inhibition, in: Metabolism of Amines in the Brain, ed. G. Hooper (MacMillan, London) pp. 38–47.

Andén, N.-E., H. Corrodi and K. Fuxe, 1971, Hallucinogenic drugs of the indolealkylamine type and central monoamine neurons, J. Pharmacol. Exptl. Therap. in press.

Bennington, F., R.D. Morin and L.C. Clark, 1965, 5-Methoxy-N,N-dimethyltryptamine, a possible endogenous psychotoxin, Alabama J. Med. Sci. 2, 397.

Carlsson, A., H. Corrodi and B. Waldeck, 1963, α-Substituierte Dopacetamide als Hemmer der Catechol-O-methyl-transferase und der enzymatischen Hydroxylierung aromatischer Aminosauren in den Catecholamin-Metabolismus eingreifende Substanzen, Helv. Chim. Acta 46, 2271.

Carlsson, A., H. Corrodi, K. Fuxe and T. Hökfelt, 1969, Effect of antidepressant drugs on the depletion of intraneuronal brain 5-hydroxytryptamine stores caused by 4-methyl-α-ethylmetatyramine, European J. Pharmacol. 5, 357.

Cerletti, A., M. Taeschler and H. Weidmann, 1968, Pharmacologic studies on the structure–activity relationship of hydroxyindole alkylamines, in: Advances in Pharmacology, eds. S. Garattini and P.S. Shore (Academic Press, New York) pp. 233–246.

Chase, T.N., G.R. Breese and I.J. Kopin, 1967, Serotonin release from brain slices by electrical stimulation: Regional differences and effect of L.S.D., Sciences 157, 1461.

Corrodi, H. and K. Fuxe, 1968, The effect of imipramine on central monoamine neurons, J. Pharm. Pharmacol. 20, 230.

Corrodi, H. and K. Fuxe, 1969, Decreased turnover in central 5-HT nerve terminals induced by antidepressant drugs of the imipramine type, European J. Pharmacol. 1, 56.

Corrodi, H. and G. Jonsson, 1965, Fluoreszenzmethoden zur histochemischen Sichtbarmachung von Monoaminen. 5. Identifizierung des fluoreszierenden Produktes aus Modellversuchen mit 5-Methoxytryptamin und Formaldehyde, Acta Histochem. (Jena) 22, 247.

Corrodi, H. and G. Jonsson, 1967, The formaldehyde fluorescence method for the histochemical demonstration of biogenic monoamines, J. Histochem. Cytochem. 15, 65.

Corrodi, H., K. Fuxe and T. Hökfelt, 1968, The effect of immobilization stress on the activity of central monoamine neurons, Life Sci. 7, 107.

Corrodi, H., K. Fuxe, P. Lidbrink and L. Olson, 1971, Minor tranquillizers, stress and central catecholamine neurons, Brain Res. 29, 1.

Diaz, P.M., S.H. Ngai and E. Costa, 1968, Factors modulating brain serotonin turnover, in: Advances in Pharmacology, eds. S. Garattini and P.A. Shore (Academic Press, New York) pp. 75–92.

Falck, B., N.-A. Hillarp, G. Thieme and A. Torp, 1962, Fluorescence of catecholamines and related compounds condensed with formaldehyde, J. Histochem. Cytochem. 10, 348.

Farnebo, L.-O. and B. Hamberger, 1971, Drug-induced changes in the release of ^{3}H-monoamines from field stimulated rat brain slices, Acta Physiol. Scand. Suppl. 371, 35.

Freedman, D.X., R. Gottlieb and R.A. Lovell, 1970, Psychotomimetic drugs and brain 5-hydroxytryptamine metabolism, Biochem. Pharmacol. 19, 1181.

Fuxe, K., 1965, Evidence for the existence of monoamine neurons in the central nervous system. IV. The distribution of monoamine nerve terminals in the central nervous system, Acta Physiol. Scand. 64, 39.

Fuxe, K. and G. Jonsson, 1967, A modification of the histochemical fluorescence method for the improved localization of 5-HT, Histochemie 11, 161.

Fuxe, K., T. Hökfelt and U. Ungerstedt, 1970a, Morphological and functional aspects of central monoamine neurons, Intern. Rev. Neurobiol. 13, 93.

Fuxe, K., T. Hökfelt, G. Jonsson and U. Ungerstedt, 1970b, Fluorescence microscopy in neuroanatomy, in: Contemporary Research Methods in Neuroanatomy, eds. W.J.H. Nauta and S.O.E. Ebbesson (Springer Verlag, Berlin, Heidelberg, New York) pp. 275–314.

Gessner, P.K., 1970, Pharmacological studies of 5-methoxy-N,N-dimethyltryptamine, LSD and other hallucinogens, in: Psychotomimetic Drugs, ed. D.H. Efron (Raven Press, New York) pp. 105–122.

Gessner, P.K. and I.H. Page, 1962, Behavioral effects of 5-methoxy-N,N-dimethyltryptamine, other tryptamines and LSD, Amer. J. Physiol. 203, 167.

Grahame-Smith, D.G., 1971, Studies in vivo on the relationship between brain tryptophan, brain 5-HT synthesis and hyperactivity in rats treated with a monoamine oxidase inhibitor and l-tryptophan, J. Neurochem. 18, 1053.

Hillarp, N.-A., K. Fuxe and A. Dahlström, 1966, Central monoamine neurons, in: Mechanisms of Release of Biogenic Amines, eds. U.S. v. Euler, S. Rosell and B. Uvnas (Pergamon Press, Oxford) pp. 31–37.

Holmstedt, B., W.J.A. Van den Heuvel, W.L. Gardiner and E.C. Horning, 1964, Separation and identification of tryptamine-related indole basis by gas chromatographic methods, Anal. Biochem. 8, 151.

Isbell, H., 1959, Comparison of the reactions induced by psilocybin and LSD-25 in man, Psychopharmacologia 1, 29.

Jones, B.E., P. Bobillier and M. Jouvet, 1969, Effet de la destruction des neurones contenant les catecholamines du mesencephale sur le cycle veille- sommeil du chat, C.R. Soc. Biol. (Paris) 163, 176.

Jonsson, G., 1969, Microfluorimetric studies of the formaldehyde-induced fluorescence of noradrenaline in adrenergic nerves of rat iris, J. Histochem. Cytochem. 17, 714.

Jonsson, G., 1971, Quantitation of biogenic monoamines demonstrated with the formaldehyde fluorescence method, Progr. Histochem. Cytochem. 2, 291.

Katz, R.I. and I.J. Kopin, 1969, Effect of D-LSD and related compounds on release of norepinephrine-H^3 and serotonin-H^3 evoked from brain slices by electrical stimulation, Pharmacol. Res. Commun. 1, 54.

Leonard, B.E. and S.R. Tonge, 1969, The effect of some hallucinogenic drugs upon the metabolism of noradrenaline, Life Sci. 8, 815.

Lidbrink, P. and G. Jonsson, 1971, Semiquantitative estimations of formaldehyde-induced fluorescence in central noradrenaline nerve terminals, J. Histochem. Cytochem. 19, 747.

Lin, R.C., S.H. Ngai and E. Costa, 1969, Lysergic acid diethylamide: role in conversion of plasma tryptophan to brain serotonin (5-hydroxytryptamine), Science 166, 237.

Mantegazzini, P., 1966, Pharmacological actions of indolealkylcamines and precursor aminoacids on the central nervous system, in: Handb. Exptl. Pharmacol. Vol. XIX, eds. O. Eichler and A. Farah (Springer Verlag, Berlin–Heidelberg–New York) pp. 424–470.

Meek, J.L. and K. Fuxe, 1971, Serotonin accumulation after monoamine oxidase inhibition, Biochem. Pharmacol. 20, 693.

Meek, J., K. Fuxe and N.-E. Andén, 1970, Effects of antidepressant drugs of the imipramine type on central 5-hydroxytryptamine neurotransmission, European J. Pharmacol. 9, 325.

Olson, L. and U. Ungerstedt, 1970, Monoamine fluorescence in CNS smears: sensitive and rapid visualization of nerve terminals without freeze-drying, Brain Res. 17, 343.

Olson, L., B. Hamberger, G. Jonsson and T. Malmfors, 1968, Combined fluorescence histochemistry and ^{3}H-noradrenaline measurements of adrenergic nerves, Histochem. 15, 38.

Persson, T., 1970, Catecholamine turnover in central nervous system, Reports from The Psychiatric Research Centre, St. Jorgen's Hospital, Goteborg.

Randic, M. and A. Padjen, 1971, Effect of N,N-dimethyltryptamine and D-lysergic acid diethylamide on the release of 5-hydroxyindoles in rat forebrain, Nature 230, 532.

Reis, D.J. and K. Fuxe, 1968, Depletion of noradrenaline in brain stem neurons during sham rage behaviour produced by acute brain stem transection in cat, Brain Res. 7, 448.

Reis, D.J. and K. Fuxe, 1969, Brain norepinephrine: Evidence that neuronal release is essential for sham rage behaviour following brain stem transection in cat, Proc. Natl. Acad. Sci. (Wash.) 64, 108.

Rosecrans, J.A., R.A. Lovell and D.X. Freedman, 1967, Effects if lysergic acid diethylamide on the metabolism of brain 5-hydroxytryptamine, Biochem. Pharmacol. 16, 2011.

Schubert, J., H. Nyback and G. Sedvall, 1970, Accumulation and disappearance of ^{3}H-5-hydroxytryptamine formed from ^{3}H-tryptophan in mouse brain: effect of LSD-25, European J. Pharmacol. 10, 215.

Sheard, M.H. and G.K. Aghajanian, 1968, Stimulation of midbrain raphe neurons: behavioral effects of serotonin release, Life Sci. 7, 19.

Sugrue, M.F., 1969, A study of the role of noradrenaline in behavioural changes produced in the rat psychotomimetic drugs, Brit. J. Pharmacol. 35, 243.

Thierry, A.M., F. Javoy, J. Glowinski and S.S. Kety, 1968, Effects of stress on the metabolism of norepinephrine, dopamine and serotonin in the central nervous system of the rat. I. Modification of norepinephrine turnover, J. Pharmacol. Exptl. Therap. 163, 163.

Udenfriend, S., 1966, Tyrosine hydroxylase, Pharmacol. Rev. 18, 43.

Wurtman, R.J. and J. Axelrod, 1963, A sensitive and specific assay for the estimation of monoamine oxidase, Biochem. Pharmacol. 12, 1439.