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Effect of chronic treatment with 5-HT_1 agonist (8-OH-DPAT and RU 24969) and antagonist (isapirone) drugs on the behavioural responses of mice to 5-HT_1 and 5-HT_2 agonists

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1 The effects of chronic (14 day) administration to mice of the 5-HT_1 agonists 8-hydroxy 2-(di-*n*-propylamino) tetralin (8-OH-DPAT) and 5-methoxy-3 (1,2,3,6-tetrahydropyridin-4-yl) IH indole (RU 24969) on the hypothermic response to 8-OH-DPAT and the locomotor response to RU 24969 have been examined.

2 Chronic administration of 8-OH-DPAT (5 mg kg^{-1}, s.c.) resulted in an attenuated hypothermic response to this drug given subcutaneously (s.c.) or intracerebroventricularly (i.c.v.) but did not alter the locomotor response to RU 24969.

3 Chronic injection of RU 24969 (3 mg kg^{-1}, i.p.) produced an attenuated locomotor response to this drug given i.p. or i.c.v. but not the hypothermic response to 8-OH-DPAT (0.5 mg kg^{-1}, s.c.).

4 Chronic administration of the putative presynaptic 5-HT_1 antagonist isapirone (10 mg kg^{-1}, i.p.) decreased the hypothermic response following 8-OH-DPAT injection but did not alter RU 24969-induced locomotion.

5 Chronic treatment with 8-OH-DPAT (5 mg kg^{-1}, s.c.) produced a modest enhancement of the 5-HT_2 receptor-mediated head-twitch behaviour initiated by 5-hydroxytryptophan injection while chronic isapirone decreased this behavioural response. 5-HT_2 receptor number in frontal cortex was unaltered by isapirone treatment but markedly decreased (34%) by chronic 8-OH-DPAT.

6 These data suggest that chronic administration of the 5-HT_1 agonists induces tolerance in their respective responses but not cross-tolerance, while chronic isapirone may down-regulate the 5-HT_{1A} site in a matter analogous to that seen by 5-HT_2 receptors following 5-HT_2 receptor antagonists.

7 The data further demonstrate that chronic treatment with 8-OH-DPAT and isapirone alter postsynaptic 5-HT_2 receptor function although 5-HT_2 receptor number in the frontal cortex did not correlate with the behavioural change.

Introduction

It is now widely acknowledged that there are several 5-hydroxytryptamine (5-HT) binding site sub-types. With regard to the 5-HT_1 sites, there is evidence for further sub-classification based primarily on ligand binding (Pedigo *et al.*, 1981; Middlemiss & Fozard, 1983). It has been suggested that 8-hydroxy 2-(di-*n*-propylamino) tetralin (8-OH-DPAT) is a specific ligand for the 5-HT_{1A} sub-type (Middlemiss & Fozard, 1983; Tricklebank *et al.*, 1984) while 5-methoxy-3 (1,2,3,6-tetrahydropyridin-4-yl) IH indole (RU 24969) shows greater selectivity for the 5-HT_{1B} site (Sills *et al.*, 1984); however, there are indications from both biochemical and behavioural studies that RU 24969 has less selectivity (Sills *et al.*, 1984; Tricklebank *et al.*, 1986). Nevertheless 8-OH-DPAT and RU 24969 produce very different functional responses in mice, the former inducing hypothermia (Goodwin *et al.*, 1985) and the latter locomotor activity (Gardner & Guy 1983; Green *et al.*, 1984).

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In addition it has recently been found that isapirone (formerly known as TVX Q 7821) is a good antagonist in mice of the hypothermic response following 8-OH-DPAT but has no effect on RU 24969-induced locomotion (Goodwin *et al.*, 1986).

The current study was undertaken for two purposes. The first was to examine whether longer term administration of the 5-HT_1 agonist drugs, 8-OH-DPAT and RU 24969, would induce a decreased sensitivity in the behavioural response to the same drug or a 'cross tolerance' to the other drug. Secondly since both 8-OH-DPAT and RU 24969 have been shown to act at presynaptic 5-HT_1 sites, whether repeated administration of the 5-HT_1 agonists or the putative 5-HT_{1A} antagonist isapirone would result in an alteration of a known postsynaptic 5-HT receptor-mediated behaviour, namely, the 5-HT_2 receptor-mediated head-twitch response (see Green & Heal, 1985 for review).

Methods

Animals

Mice were male C57/Bl/6/Ola (Olac, Bicester) weighing 22–28 g at the time of study, housed in groups of 8–10 and given modified 41B pellets and tap water *ad libitum*. They were kept in conditions of controlled temperature (20°C + 1°C) and lighting (dark period 19 h 00 min–07 h 00 min).

Behavioural studies

Head-twitch behaviour was studied after agonist administration. Mice were injected with 5-methoxy-*N,N*-dimethyltryptamine (5-MeODMT; 5 mg kg^{-1}, i.p.) and the total number of head-twitches measured in the next 6 min. The observer was unaware of the nature of the pretreatment drug.

Locomotor activity was measured simultaneously for pairs of control and experimental mice with LKB Animex activity meters (sensitivity and tuning; 30 μA).

Temperature measurement

Body temperature was measured with a Comark thermocouple with digital read-out and rectal probe inserted 2.5 cm into the colon, the animal being lightly restrained in the hand during measurement. The probe was lubricated with handcream before each use.

Measurement of brain 5-hydroxytryptamine

Whole mouse brain 5-HT was measured fluorimetrically by the method of Curzon & Green (1970) and 5-HT synthesis estimated by measurement of the accumulation of 5-HT following monoamine oxidase inhibition with tranylcypromine (Neff & Tozer, 1968).

Intracerebroventricular injection techniques

Drugs were dissolved in saline and injected in a total volume of 2 μl intracerebroventricularly as described in detail elsewhere (Heal, 1984). Animals were briefly anaesthetized with halothane (ICI Pharmaceuticals) during the procedure.

5-HT_2 receptor binding studies

The number of 5-HT_2 receptors in frontal cortex was measured in pooled tissue from 5 animals with [^{3}H]-ketanserin (specific activity 76.7 Ci $mmol^{-1}$, N.E.N.) using a slight modification of the method of Leysen *et al.* (1982). Non specific binding was determined by displacement with methysergide (10^{-6} M). Scatchard analysis of data was performed following incubation with 6 concentrations of [^{3}H]-ketanserin (0.24–4.86 nM) using linear regression analysis by the method of least squares ($r = 0.92$ or better in all cases). Tissue protein concentration was determined by the method of Lowry *et al.* (1951).

Drugs

Drugs were obtained from the following sources (in parentheses): 5-methoxy-*N,N*-dimethyltryptamine (Sigma, Poole); 8-OH-DPAT (Research Biochemicals Inc. Wayland, MA, U.S.A.); tranylcypromine (Smith, Kline and French, Welwyn Garden City); RU 24969 (gift from T.P. Blackburn, ICI Pharmaceuticals); isapirone (Troponwerke, Cologne). All drugs were dissolved in 0.9% NaCl (saline) and injected i.p. except where stated otherwise.

Statistics

Locomotor responses and temperature changes are reported as mean ± s.e.mean and have been analysed by Student's *t* test (unpaired). All other behavioural scores were analysed with the Mann Whitney rank order test for non parametric data.

Results

Changes in the hypothermic response to 8-OH-DPAT during chronic treatment with the drug

Mice were injected with a high dose of 8-OH-DPAT (5 mg kg^{-1} s.c.) and the rectal temperature measured over the next 90 min. A sustained temperature decrease was induced by the 8-OH-DPAT (Figure 1). The 8-OH-DPAT-injected mice were then injected

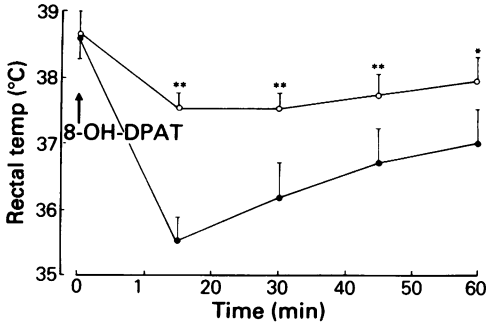

Figure 1 Effect of chronic treatment with 8-OH-DPAT on the hypothermic response to 8-OH-DPAT. Mice were injected with 8-OH-DPAT (5 mg kg^{-1}, s.c.) and the temperature change recorded on day 1 (●) and after 12 further injections on day 14 (○). The chronically treated mice showed an attenuated response on day 14 compared with day 1 at all time points following injection: $*P < 0.005$; $**P < 0.001$. There was no difference in the basal temperature between the two groups.

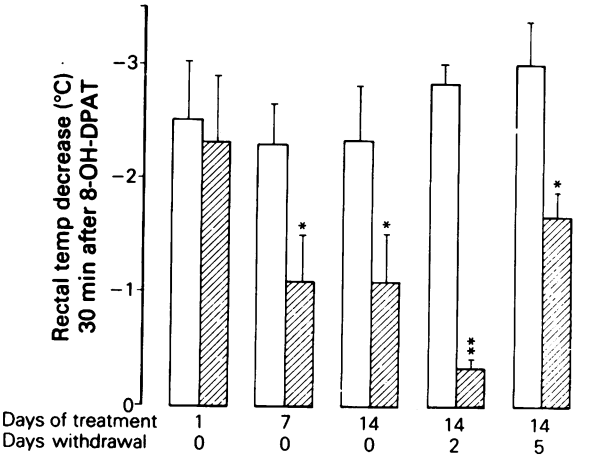

Figure 2 Time course of the attenuation of the temperature decrease induced by 8-OH-DPAT following chronic treatment with 8-OH-DPAT. Mice were injected for up to 14 days with 8-OH-DPAT (5 mg kg^{-1}) and the temperature decrease measured on the first, seventh and fourteenth injection and 2 and 5 days after the last injection. Results show mean temperature decrease + s.d. (bar) of the saline (open column) and 8-OH-DPAT (hatched column) treated mice. Different from saline infected: $*P < 0.01$; $**P < 0.001$. There were no differences in the basal temperature between the groups.

once daily with 8-OH-DPAT for a further 13 days. The temperature decrease induced by the drug was measured following the injection on day 14. The temperature response was attenuated (Figure 1) compared with day 1, suggesting that subsensitivity of the 8-OH-DPAT site had been induced.

In the next study mice were injected on day 1 with saline or 8-OH-DPAT (5 mg kg^{-1}, s.c.) and the injections repeated once daily for 6 or 13 days. On day 7 and 14 both groups were injected with 8-OH-DPAT (5 mg kg^{-1}). The repeated saline injections did not alter the degree of hypothermia induced by the 8-OH-DPAT injection (Figure 2) in contrast to the chronic 8-OH-DPAT-treated mice (Figure 2) where subsensitivity could be seen to have developed even by day 7.

Further groups of mice treated chronically with 8-OH-DPAT were withdrawn for 2 and 5 days from treatment and then tested with 8-OH-DPAT (5 mg kg^{-1}). The attenuation of the hypothermia was as pronounced after 2 days withdrawal but less marked after 5 days withdrawal (Figure 2).

To investigate whether the attenuation of the 8-OH-DPAT-induced hypothermia was due to changes in the pharmacokinetics or peripheral metabolism of the drug following its chronic administration, mice were treated chronically with saline or 8-OH-DPAT (5 mg kg^{-1}) for 14 days and the temperature decrease induced by intracerebroventricular injection of 8-OH-DPAT (2 μg in 2 μl) measured. The mice injected chronically with 8-OH-DPAT showed a smaller temperature decrease than chronic saline-injected animals at the nadir of the response (Figure 3).

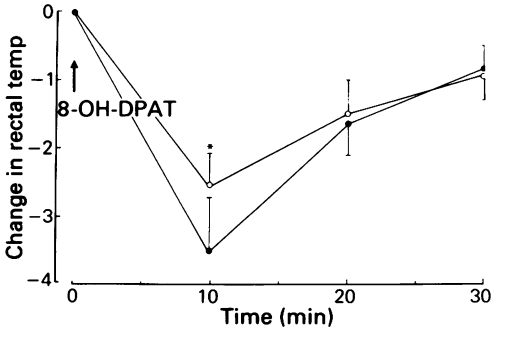

Figure 3 The hypothermic response of mice following 8-OH-DPAT (2 μg in 3 μl) injected intracerebroventricularly. Mice were injected daily for 14 days with either 8-OH-DPAT (5 mg kg^{-1}, s.c.) or saline and on day 15 both the chronic 8-OH-DPAT- (○) and saline (●)-treated animals were injected with 8-OH-DPAT (i.c.v.). Responses show mean + s.d. (as bar) of both groups with an attenuation seen 10 min after injection ($*P < 0.01$).

Effect of chronic 8-OH-DPAT injection on the locomotor response produced by RU 24969 injection

Mice were injected with RU 24969 (3 mg kg^{-1}, s.c.) and the resultant locomotor activity measured over the next 60 min. Chronic treatment with 8-OH-DPAT did not alter the locomotor response to RU 24969 compared with chronic saline-injected control mice (Table 1).

Effect of chronic treatment with RU 24969 on the hypothermic response to 8-OH-DPAT

Mice were injected once daily with either saline or RU 24969 (3 mg kg^{-1}, i.p.) for 14 days. Twenty-four hours after the last injection the temperature change induced by a low dose of 8-OH-DPAT (0.5 mg kg^{-1}, s.c.) was measured. The temperature decrease produced by 8-OH-DPAT was similar in the chronic RU 24969- and saline-injected groups (Figure 4).

Effect of chronic administration with RU 24969 on the locomotor response to RU 24969

Repeated administration of RU 24969 (3 mg kg^{-1}) for 14 days reduced the locomotor response of the mice to this drug (Table 1). To investigate whether this was due to altered metabolism of the drug, we next examined the effect of repeated treatment on the response of mice to RU 24969 injected intracerebroventricularly. In order to do this it was first necessary to establish that RU 24969 injected i.c.v. produced locomotion and the dose-response curve of this effect. Intracerebroventricular injection of RU 24969 (10–500 ng in 2 µl saline) produced a dose-dependent increase in behavioural activity and locomotion (Figure 5a) with an approximate ED_{50} of 20 ng. The mice displayed co-ordinated locomotor activity which was intermittent and random. The locomotion was performed with a markedly hunched body posture with head pulled in and abdomen held clear of the floor. During the periods of immobility there was often intense grooming. Other behavioural changes observed were almost constant sniffing and head bobbing as well as frequent rearing. Pilo-erection occurred and occasional single head-twitches.

Groups of mice were next treated for 14 days with intraperitoneal injections of either saline (control group) or RU 24969 (3 mg kg^{-1}). Twenty-four hours after the last dose both groups were injected with RU 24969 (50 ng in 2 µl i.c.v.). Both groups showed all the behavioural changes and locomotor changes described above. However the intensity of the response was markedly reduced in the mice previously treated repeatedly with RU 24969 peripherally (Figure 5b).

Effect of chronic isapirone on the hypothermic response of mice to 8-OH-DPAT

Mice were injected daily for 14 days with saline or isapirone (10 mg kg^{-1}) and the hypothermic response to 8-OH-DPAT (0.5 mg kg^{-1} s.c.) examined 24 h after the last treatment.

Mice treated chronically with isapirone showed an attenuated hypothermic response both 24 h and 72 h after the last injection (Figure 4).

Effect of chronic isapirone on the locomotor response following RU 24969

The locomotor response induced by RU 24969 (3 mg kg^{-1}) was unaltered by 14 days treatment with isapirone (10 mg kg^{-1}) (Table 1).

Effect of chronic 8-OH-DPAT, RU 24969 and isapirone on the head twitch response induced by 5-MeODMT

Mice were injected daily for 14 days with saline, 8-OH-DPAT (5 mg kg^{-1}). RU 24969 (3 mg kg^{-1}) or isapirone (10 mg kg^{-1}) and 24 h after the last dose the head twitch response to 5-MeODMT (5 mg kg^{-1}) was examined. Because of the between-experiment variation, control groups (chronic saline) were always run and tested at the same time as the experimental group.

Chronic treatment with RU 24969 did not alter the 5-MeODMT-induced head twitch response (Figure 6). Chronic treatment with 8-OH-DPAT produced a modest but statistically significant enhancement of the head-twitch response (Figure 6), while chronic treatment with isapirone produced a modest but significant inhibition of the response (Figure 6).

Table 1 Locomotor response following RU 24969 injection after chronic administration of isapirone, 8-OH-DPAT and RU 24969

Injected (i.p.)	*Locomotor activity* (automated counts per 60 min)
Saline	7501 ± 756 (9)
Isapirone	7458 ± 983 (5)
8-OH-DPAT	6844 ± 1067 (5)
RU 24969	3906 ± 1839 (9)*

Results show counts per 60 min of pairs of mice following injection of RU 24969 (3 mg kg^{-1}). Results expressed as mean ± s.d. with number of experiments in parentheses. Locomotor responses of mice following saline were 638 ± 212 (4). *Different from saline-injected mice: $P < 0.001$.

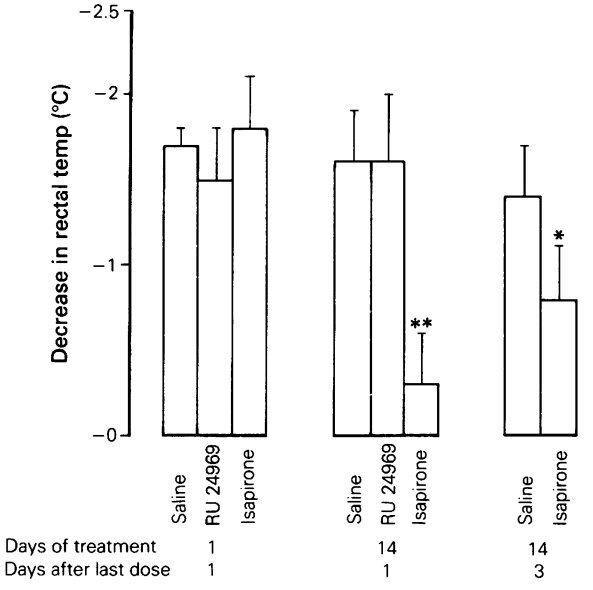

Figure 4 Effect of chronic treatment with RU 24969 (5 mg kg^{-1}) and isapirone (10 mg kg^{-1}) on the hypothermic response to 8-OH-DPAT (0.5 mg kg^{-1}, s.c.). Mice were treated with the drugs for 1 or 14 days and tested either 1 or 3 days later. Responses shown as mean and ± s.d. shown as bar. Isapirone-treated rats different from saline-injected controls: $*P < 0.01$; $**P < 0.001$.

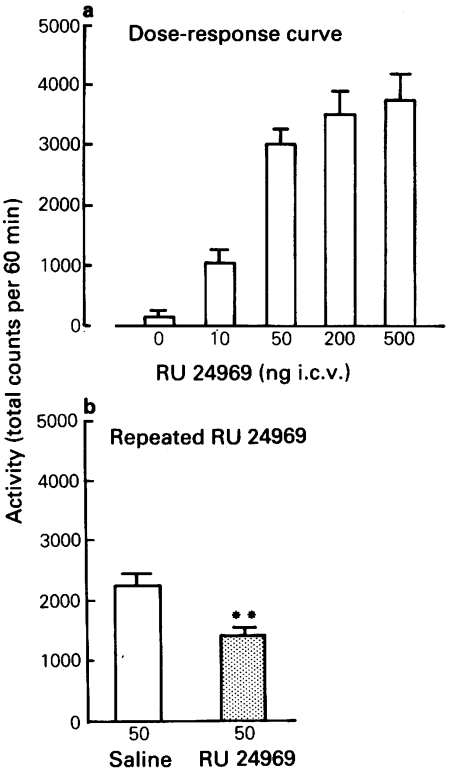

Figure 5 (a) Effect of increasing doses of RU 24969 injected i.c.v. on the locomotor activity of mice ($n = 4$ for each dose). (b) The locomotor response of mice to RU 24969 (50 ng i.c.v.) following repeated treatment with either saline or RU 24969 (3 mg kg^{-1} daily). Repeated RU 24969 treated mice significantly different from saline-injected animals: $**P < 0.01$.

Effect of chronic treatment with 8-OH-DPAT, and isapirone on 5-HT$_2$ receptor number in frontal cortex

Previous studies on the consequences of antidepressant drug and administration on head-twitch response had shown a reasonable correlation between head-twitch response and 5-HT$_2$ receptor density in frontal cortex (Goodwin *et al.*, 1984) although a good relationship was not found following lithium administration (Goodwin *et al.*, 1986). Since chronic 8-OH-DPAT and isapirone injection altered head-twitch behaviour we next examined 5-HT$_2$ receptor density in the frontal cortex following 14 days treatment with both drugs.

No change in receptor density (B_{max}) or dissociation constant (K_D) was seen following isapirone treatment (Table 2) while surprisingly, chronic treatment with 8-OH-DPAT actually decreased 5-HT$_2$ receptor number (Table 2).

Effect of chronic treatment with 8-OH-DPAT, RU 24969 and isapirone on the synthesis rate of 5-HT in whole brain

Acute administration of both 8-OH-DPAT and RU

Table 2 Effect of chronic isapirone and 8-OH-DPAT administration on 5-HT$_2$ receptor binding characteristics in frontal cortex of mice

Injected	*5-HT$_2$ binding characteristics* B_{max}	K_D	*n*
Saline	426 ± 58	0.83 ± 0.17	3
Isapirone	445 ± 47	0.92 ± 0.39	3
8-OH-DPAT	280 ± 35*	0.70 ± 0.08	3

Results reported as mean ± s.d. Saline and both drugs given once daily, isapirone at 10 mg kg^{-1} and 8-OH-DPAT at 5 mg kg^{-1}. B_{max} expressed in fmol-mg^{-1} protein and K_D in nM.
*Different from control value: $P < 0.01$.

24969 decreases the rate of 5-HT synthesis in the brain (Goodwin & Green, 1985) while isapirone injection produces a modest enhancement of the rate (Goodwin *et al.*, 1986). The rate of synthesis was measured in mouse brain 24 h after the last of 14 once daily injections of the 3 drugs. Chronic treatment with either isapirone or RU 24969 produced no change in brain 5-HT content or rate of 5-HT synthesis (Table 3). Mice treated with 8-OH-DPAT had a lowered concentration of 5-HT in the brain and a lowered rate of 5-HT synthesis (Table 3).

Discussion

The current study was conducted in mice and it is worth, at the outset, outlining the advantages of mice over rats in this type of investigation. Firstly there is good evidence that the hypothermic response in mice which follows 8-OH-DPAT injection is mediated by 5-HT_{1A} receptors located presynaptically (Goodwin *et al.*, 1985). By contrast, in rats, in addition to the hypothermic response there are also behavioural responses to 8-OH-DPAT and the latter probably result from stimulation of postsynaptic 5-HT_{1A} receptors (Goodwin & Green 1985; Goodwin, De Souza & Green, unpublished). No obvious postsynaptic 5-HT_{1A}-mediated response following 8-OH-DPAT injection to mice has been observed.

Secondly, in mice, isapirone appears to act as a 5-HT_{1A} antagonist, totally inhibiting the 8-OH-DPAT-induced hypothermia (Goodwin *et al.*, 1986). In rats, studies suggest that the drugs act as a mixed agonist/antagonist (Goodwin *et al.*, 1986; Cunningham *et al.*, 1985).

Finally, the mouse displays a simple easily quantified 5-HT_2 receptor-mediated behavioural response, namely the head-twitch response. For all these reasons therefore mice were chosen in the hope that results obtained would be easier to interpret.

One of the objects of this investigation was to try and determine the specificity of the 5-HT_1 drugs for the possible 5-HT_1 receptor sub-types. Repeated administration of drugs that are either agonist or antagonist at neurotransmitter receptors has been found in a variety of studies to alter the sensitivity of that receptor. Since ligand binding studies have suggested that RU 24969 might have some activity at the 5-HT_{1A} site (Sills *et al.*, 1984; Tricklebank *et al.*, 1984) and isapirone appears, at least in rats (see above), to have both antagonist and agonist properties at the 5-HT_{1A} site (Cunningham *et al.*, 1985; Goodwin *et al.*, 1986) it was thought that 'cross sensitivity' might be demonstrable between the drugs used and their sites of action.

The major changes seen have been summarized in Table 4. Briefly, 8-OH-DPAT and RU 24969 induced subsensitivity in their respective sites but did not show cross sensitivity and repeated administration of drugs acting at 5-HT_1 sites altered the response to drugs acting at the 5-HT_2 site. These changes will now be discussed in detail.

To deal with 8-OH-DPAT first: chronic administration of this drug induced a decrease of the sensitivity of the hypothermia response. This decrease in the response is attributable to a pharmacodynamic subsensitivity and not a change in the metabolism of the drug since subsensitivity was seen at the time of maximum change in the response to the drug injected i.c.v. and animals treated chronically with 8-OH-DPAT also showed a decrease in the hypothermic response to gepirone, another drug with agonist actions at the 5-HT_{1A} receptor (Goodwin & Green, unpublished).

In contrast, chronic 8-OH-DPAT administration in no way altered the behavioural response of the animals to injection of RU 24969

Similarly RU 24969 given repeatedly decreased the response of the animals to that drug but not their response to 8-OH-DPAT. Again the indications from the experiments giving RU 24969 i.c.v. are that a true receptor-mediated subsensitivity had been induced.

Together these experiments suggest that repeated administration of 8-OH-DPAT or RU 24969 induces subsensitivity in the respective 5-HT_1 receptor subtype at which the drug has been predominantly suggested to act but not the other 5-HT_1 receptor subtype.

Table 3 Concentration of 5-HT and the rate of 5-HT synthesis in mouse brain following chronic treatment with isapirone, RU 24969 and 8-OH-DPAT

Injected	*Brain 5-HT concentration* Saline	*Tranylcypromine*	*Synthesis rate*
Saline	0.42 ± 0.05 (5)	0.59 ± 0.05 (5)	0.17
Isapirone	0.41 ± 0.03 (5)	0.56 ± 0.08 (5)	0.15
RU 24969	0.41 ± 0.03 (6)	0.59 ± 0.06 (6)	0.18
8-OH-DPAT	0.33 ± 0.01 (6)*	0.44 ± 0.04 (6)**	0.11

Mice were injected daily with saline, isapirone (10 mg kg^{-1}), RU 24969 (3 mg kg^{-1}) or 8-OH-DPAT (5 mg kg^{-}) for 14 days as described in Methods. Brain 5-HT concentration was determined 24 h after the last treatment. Synthesis was calculated by measurement of the accumulation of 5-HT in the 60 min following the MAO inhibitor tranylcypromine (5 mg kg^{-1}). Concentration is expressed in μg 5-HT g^{-1} brain (wet weight) and synthesis in μg 5-HT g^{-1} brain (wet weight) per hour. Different from saline injected controls: $*P<0.005$; $**P<0.001$.

Table 4 Summary of major behavioural changes following repeated treatment with drugs acting at 5-HT$_1$ receptors

Chronic treatment with	*8-OH-DPAT-induced hypothermia*	*RU 24969-induced locomotion*	*5-HT$_2$-mediated head-twitch*
8-OH-DPAT	↓	○	↑
RU 42969	○	↓	○
Isapirone	↓	○	↓

Arrows indicate direction of change compared with response seen in control animals: ↓ diminished response; ↑ enhanced response; ○ no change.

Isapirone given repeatedly did not alter the locomotor response to RU 24969 but did markedly attenuate the hypothermic response following 8-OH-DPAT. Isapirone attenuates this response in mice when given before 8-OH-DPAT, suggesting that it acts as a 5-HT$_{1A}$ antagonist (Goodwin *et al.*, 1986). It seems unlikely however that the change seen was merely due to accumulation of the drug on repeated administration since the isapirone seems to have a short half-life in mice (less than 6 h; Goodwin *et al.*, 1986). Furthermore, an attenuation was still seen 72 h after the last injection of the antagonist drug. It is more likely that the 5-HT$_{1A}$ site shows a down regulation on antagonist administration in a manner analogous to the 5-HT$_2$ receptor (Blackshear *et al.*, 1983; Leysen *et al.*, 1986).

This study has also demonstrated that manipulation of presynaptic 5-HT receptors does alter the sensitivity of postsynaptic 5-HT$_2$ receptors. Indications from electrophysiological studies suggest that 8-OH-DPAT acts at somato-dendritic receptors rather than terminal sites (Sprouse & Aghajanian, 1985). Repeated stimulation of these 5-HT$_{1A}$ sites (presumably shutting down 5-HT synthesis and release) resulted in an increase in 5-HT$_2$ receptor-mediated head-twitch behaviour while continued antagonism of the 5-HT$_{1A}$ site (possibly leading to enhanced release) produced a decrease in the 5-HT$_2$-mediated response. The changes in the head-twitch response were not reflected in the 5-HT$_2$ receptor density in the frontal cortex. However since head twitch behaviour probably originates in the hind-brain (see Green & Heal, 1985) this is not totally unexpected. What was surprising was that 5-HT$_2$ density actually decreased after repeated 8-OH-DPAT, possibly indicating regional variation in the changes occurring in terminal regions following injection of this drug.

Only repeated 8-OH-DPAT injection produced a change in 5-HT synthesis rate in whole brain. The lowered brain content of 5-HT meant that the 'turnover time' (that is, the time taken to replace the amine

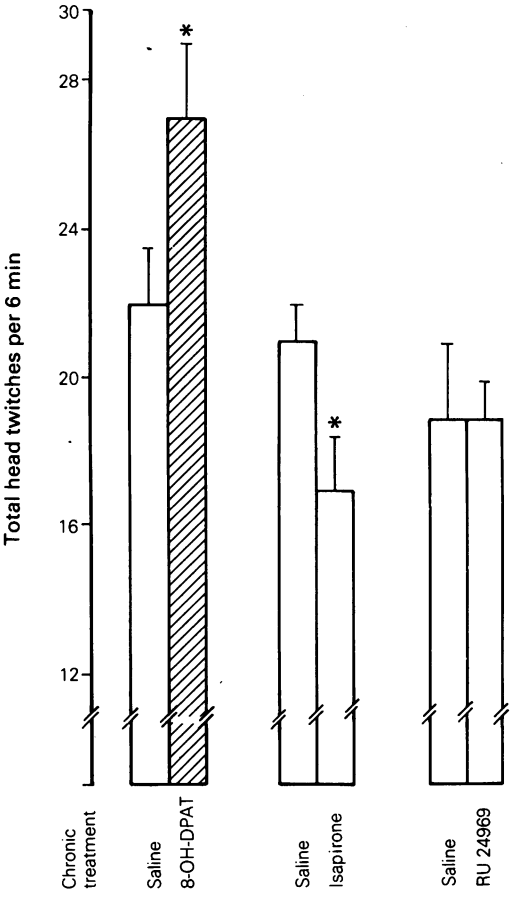

Figure 6 Head twitch response induced by 5-MeODMT (5 mg kg^{-1}) injection in mice treated for 14 days with 8-OH-DPAT (5 mg kg^{-1} s.c.), isapirone (10 mg kg^{-1}) and RU 24969 (5 mg kg^{-1}). Behavioural tests performed 24 h after the last dose of the drugs given chronically. Results show median and bars interquartile ranges. Different from saline-injected controls: $^*P < 0.05$.

pool and which is the product of synthesis rate and brain content) was little changed. These data should be extended to a study of brain regions, given the results obtained with head-twitch response and 5-HT_2 binding.

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