Potential anxiolytic properties of R-(+)-8-OSO$_2$CF$_3$-PAT, a 5-HT$_{1A}$ receptor agonist

Tjeerd Barf [a,*], S. Mechiel Korte [b], Gerdien Korte-Bouws [b], Clas Sonesson [c], Geert Damsma [a,1], Béla Bohus [b], Håkan Wikström [a]

[a] Department of Medicinal Chemistry, University Centre for Pharmacy. A. Deusinglaan 2. NL-9713 AW Groningen, Netherlands
[b] Department of Animal Physiology, University of Groningen, Biological Centre, P.O. Box 14, NL-9750 AA Haren, Netherlands
[c] Medicinal Chemistry Unit at the Department of Pharmacology, University of Göteborg, Medicinaregatan 7, S-413 90 Göteborg, Sweden

Abstract

The anxiolytic property of R-(+)-8-OSO$_2$CF$_3$-PAT (R-(+)-8-[[(trifluoromethyl)sulfonyl]oxy]-2-(n-propyl-amino)tetralin), a 5-HT$_{1A}$ receptor agonist, was evaluated in Wistar rats by means of animal models of anxiety, the conditioned defensive burying model and the conditioned stress-induced freezing response followed by the elevated plus-maze test, respectively. In addition, the 5-HIAA/5-HT ratio (5-hydroxy-indole acetic acid/5-hydroxytryptamine) of rat brain homogenates was studied. Acute drug administration resulted in abolition of the burying behaviour (3 mg/kg i.p.), a dose-dependent decrease of rearing and induction of hyperphagia. R-(+)-8-OSO$_2$CF$_3$-PAT had no effect on conditioned footshock-induced freezing behaviour but increased open-arm activity in the rats on the plus-maze. The 5-HIAA/5-HT ratio was decreased in the lateral septum (1 and 3 mg/kg), dorsal hippocampus (3 mg/kg) and somatosensory cortex (3 mg/kg), implying that R-(+)-8-OSO$_2$CF$_3$-PAT affects particularly the limbic system in anxiety-inducing situations.

Keywords: 5-HT$_{1A}$ receptor; Defensive burying; Inescapable footshock; Plus-maze; 5-HIAA/5-HT (5-hydroxy-indole acetic acid/5-hydroxytryptamine) ratio

1. Introduction

A number of studies have shown that drugs that reduce serotonin (5-hydroxytryptamine; 5-HT) function produce anxiolytic-like effects. Acute administration of 5-HT$_{1A}$ receptor ligands such as 8-hydroxy-2-(di-n-propyl-amino)tetralin (8-OH-DPAT; Fig. 1) are thought to reduce serotonergic function by acting as agonists at somatodendritic autoreceptors, which inhibit the firing of 5-HT neurones in raphe nuclei (Dourish et al., 1986). It is important that direct infusion of these 5-HT$_{1A}$ receptor agonists into the dorsal raphe gives reproducible anxiolytic effects, supporting this hypothesis (Higgins et al., 1988, 1992; Hogg et al., 1994).

Previous investigations revealed that R-(+)-8-[[(triflu-oromethyl)sulfonyl]oxy]-2-(n-propyl-amino)tetralin (R-(+)-8-OSO$_2$CF$_3$-PAT; Fig. 1), an analogue of 8-OH-DPAT, is a chemically and biologically stable compound possessing the pharmacological profile of a 5-HT$_{1A}$ receptor agonist (Sonesson et al., 1993, 1995). [2]

The behavioural pharmacology of the drug has not been explored as yet. Therefore, the effects of R-(+)-8-OSO$_2$CF$_3$-PAT were studied in different animal models of anxiety. In the elevated plus-maze test, rats restrict their activity to the enclosed areas, avoiding the two open arms (Pellow et al., 1985). This behaviour can be reversed and enhanced by anxiolytic and anxiogenic drugs, respectively. The plus-maze model is relatively insensitive to drugs other than anxiolytics and anxiogenics (Handley and McBlane, 1993a). Prior exposure to an emotional stressor

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* Corresponding author.
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[2] K_i values (nM): 1.3 (5-HT$_{1A}$); 6.7 (5-HT$_{1D\alpha}$); 138 (5-HT$_{1D\beta}$); 69 (D$_2$, ag) 311 (D$_2$, antag); 524 (D$_3$).
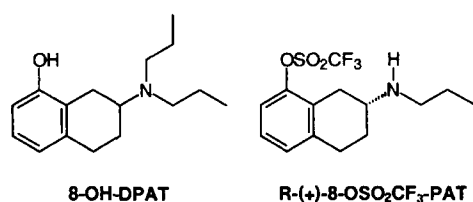
Fig. 1. Chemical structures of 8-OH-DPAT and R-(+)-8-OSO_2CF_3-PAT.

produces higher anxiety in the animals, which is reflected by reduced exploration of the open maze arms in favour of the enclosed maze arms (Korte et al., 1995).

A second animal model of anxiety is the 'conditioned defensive burying test' (Treit et al., 1981). In this test, rats are exposed to an electrified shock probe, and the duration of burying behaviour is the major index of anxiety. Standard antianxiety agents suppress the burying response in a dose-related manner.

It has been suggested that acute administration of partial agonists such as buspirone (Vandermaelen et al., 1986), ipsapirone (Sprouse and Aghajanian, 1987) and gepirone (Blier and De Montigny, 1987) decreased 5-HT turnover or lowered levels of 5-hydroxy-indole acetic acid (5-HIAA), the major metabolite of 5-HT, which may be a feature of particular brain regions. 5-HIAA/5-HT ratios are indicative of changes in the 5-HT turnover rate (Blanchard et al., 1993). Because of the expected 5-HT_{1A} agonistic property of R-(+)-8-OSO_2CF_3-PAT it was of interest to examine the 5-HIAA/5-HT ratios in several brain regions.

In the present study, the ability of R-(+)-8-OSO_2CF_3-PAT to produce anxiolytic effects after acute administration was evaluated in these animal models and from the 5-HIAA/5-HT ratio of rat brain homogenates.

2. Materials and methods

2.1. Animals

Male Wistar rats weighing 300–495 g at the beginning of the experiments were used. They were housed individually in transparent Plexiglas cages (25 × 25 × 30 cm) with a 12-h light-dark regimen (light on between 08:00–20:00 h). All animals had free access to standard rat chow (Hope Farms) and tap water. The experiments were carried out between 10:00–14:00 h.

2.2. Drugs

R-(+)-8-OSO_2CF_3-PAT was synthesized in the Department of Medicinal Chemistry, University Centre for Pharmacy in Groningen, Netherlands. The compound was dissolved in saline and given intraperitoneally (i.p.), 30 min

before the test session in a dose range of 1–3 mg/kg. Cited doses refer to the HCl salt and do not produce the 5-HT syndrome in the rats (Sonesson et al., 1995). The control group received saline.

2.3. Conditioned defensive burying

The shock-probe defensive burying test was performed in the animals' home cage. The floor was covered with wood shavings (height 2 cm). A removable Teflon probe (10 cm long, 1 cm in diameter) was positioned 2 cm above the bedding. The probe was inserted through a small hole in the center of the wall of the cage. Two exposed wires (0.5 mm in diameter) were each wrapped (25 times) independently around the probe. Whenever the animal touched both wires simultaneously with some part of its body an electric current of 1.5 mA was delivered to the animal. During the entire period the shock circuit was left on, i.e. the 'repeated shock probe procedure' was used (Treit and Fundytus, 1988). Shock intensity was adjusted with a variable resistor in series with a 1000-V shock source. On day 2 vehicle or drug was injected 30 min before the introduction of the non-electrified probe in the home cages of the rats. Thus the procedure investigated the conditioned emotional consequence of former punishment rather than the direct effect of shock. All animals were observed for 10 min. To avoid false negative results, only animals burying for more than 25% of total time on day 1 were tested for day 2.

2.4. Conditioned fear of footshock

The rats were exposed to a dark compartment (50 × 50 × 50 cm) equipped with a grid floor (Korte and Bohus, 1990), where they were allowed to stay for 5 min. During the trial an inescapable scrambled footshock (0.6 mA, AC for 3 s) was given after the 1st and the 4th minute. On the next day the animals were re-exposed to the dark footshock compartment in which no further shock was given.

2.5. Elevated plus-maze

Directly after the 5-min re-exposure to the shock compartment the animals were placed in the elevated plus-maze. The apparatus consisted of two open (50 × 10 cm) and two enclosed arms (50 × 10 × 40 cm), arranged so that the two arms of each type were opposite to each other (Pellow et al., 1985), connected by an open central area (10 × 10 cm). The maze was elevated to a height of 50 cm. Light intensity in the open arms and closed arms was 200–350 Lux and < 1 Lux, respectively. The rats were placed individually in the centre of the maze facing one of the enclosed arms. Each rat was tested for 5 min on the elevated plus-maze. The maze was cleaned after each rat had occupied it.

2.6. Behavioural measurements

The behaviour in the defensive burying model was classified in five categories: (a) defensive burying – moving toward the probe and spraying or pushing bedding material toward the probe with rapid movements of the snout or forepaws as described by Pinel and Treit (1983); (b) eating – chewing chow or faeces; (c) rearing – standing or sitting on hindlegs, mostly making sniffing movements, with the nose up into the air; (d) resting – the rat's hindlimbs, forelimbs, and belly touched the floor and supported its weight; (e) exploring – investigation of any part of the home cage. During the exposure to the former shock compartment, time spent immobile – i.e. animal completely motionless – was measured. On the elevated plus-maze, the activity scores – i.e. number of open- and closed-arm entries – were used as indices of the anxiolytic or anxiogenic effects (Pellow et al., 1985). The observations were recorded by trained observers who were blind to the treatment order.

2.7. Measurement of 5-HT and 5-HIAA

The rats were anaesthetized in an ether chamber and killed by rapid decapitation, 30 min after injection. The brains were immediately frozen in a dry ice precooled tube containing n-heptane and stored at $-70°C$ until the assays were performed. For the assay, a brain was cut in slices of 2 mm (homemade brain-slicer) at $-4°C$ after which particular brain areas were punched out on a frozen surface. The tissue samples were homogenized in icewater in a 150-μl solution containing 5 μM clorgyline, 5 μg/ml glutathione and 20 ng/ml N-ω-methylserotonin (internal standard) with a High Intensity Ultrasonic Processor (Sonics and Materials, USA). Thereafter, 12.5 μl 2 M $HClO_4$ and 10 μl 2.5 M potassium acetate were added to 50 μl of the homogenate. After 15 min the tissue samples were centrifuged for 10 min at $15000 \times g$ ($-10°C$). Thereafter, 30 μl of the supernatant was diluted with 450 μl UP.

The samples were injected onto a reverse-phase/ion-pair high performance liquid chromatography (HPLC) setup with electrochemical detection for the measurement of 5-HT and 5-HIAA. The chromatographic system consisted of an LKB 2150 HPLC pump (Pharmacia, Sweden), a Promis II autosampler (Spark, Netherlands) with a 100-μl loop and a column (150 mm $\times$ 4.6 mm i.d.) packed with Hypersil ODS, 5 μm particle size (Alltech Associates, USA).

The mobile phase consisted of 0.051 M citric acid monohydrate, 0.063 M $NaH_2PO_4 \cdot 2H_2O$, 0.403 mM EDTA, 0.356 mM sodium octyl sulphonate, 0.265 mM di-N,N-n-butylamine and 13% methanol. This buffer was set to pH 3.8 with 1 M HCl and then filtered through a 0.22-μm membrane filter (Schleicher & Schuell, Germany). Separation was done at room temperature using a flow rate of 1 ml/min.

Detection of the 5-HT and 5-HIAA was performed using an electrochemical detector (Antec, Leiden, Netherlands) with a glassy carbon working electrode set at -0.75 V (2 nA/V) versus an Ag/AgCl reference electrode. The data were recorded with a chart recorder (Model BD41, Kipp and Zn., Netherlands), and peak heights of samples were compared with those of standards determined each day for quantification. The limit of detection (signal/noise ratio 3 : 1) was 9.5 fmol/100 μl.

2.8. Statistics

The data were analyzed with a one-way analysis of variance (ANOVA). The ANOVA was followed by Dunnett's t-test in order to compare the vehicle group to each of the drug groups.

3. Results

3.1. Conditioned defensive burying

Fig. 2A shows that R-(+)-8-OSO_2CF_3-PAT dose dependently reduced the burying behaviour until complete abolition at the 3-mg/kg dose ($F(2,18) = 3.46$; $P \leq 0.05$). The time spent rearing was also significantly decreased, both at the 1-mg/kg ($F(2,18) = 10.95$; $P \leq 0.05$) and 3-mg/kg dose ($P \leq 0.01$), as depicted in Fig. 2B. Interestingly, food intake was dramatically increased at the doses applied (Fig. 2C).

3.2. Conditioned fear of footshock

The percentage of time spent immobile in the inescapable footshock compartment was not significantly affected either by the 1-mg/kg or by the 3-mg/kg dose (Fig. 3).

3.3. Elevated plus-maze

The percentage time L/L + D (L = time spent in open arms; D = time spent in enclosed arms) showed a non-significant trend towards increased open-arm activity after increasing of the dose (Fig. 4A). One-way ANOVA revealed a significant effect of R-(+)-8-OSO_2CF_3-PAT on the number of open-arm entries at 3 mg/kg ($F(2,21) = 15.27$; $P \leq 0.01$), as shown in Fig. 4B. In addition, the numbers of open-arm entries for the non-stressed and stressed animals were significantly different ($P \leq 0.05$; t-test). Fig. 4C shows that the number of enclosed-arm entries was not significantly changed at the doses applied.

3.4. Effect of acute administration of R-(+)-8-OSO₂CF₃-PAT on the 5-HIAA/5-HT ratio in various rat brain regions

Administration of 3 mg/kg of R-(+)-8-OSO_2CF_3-PAT i.p. reduced the 5-HIAA/5-HT ratio significantly in the
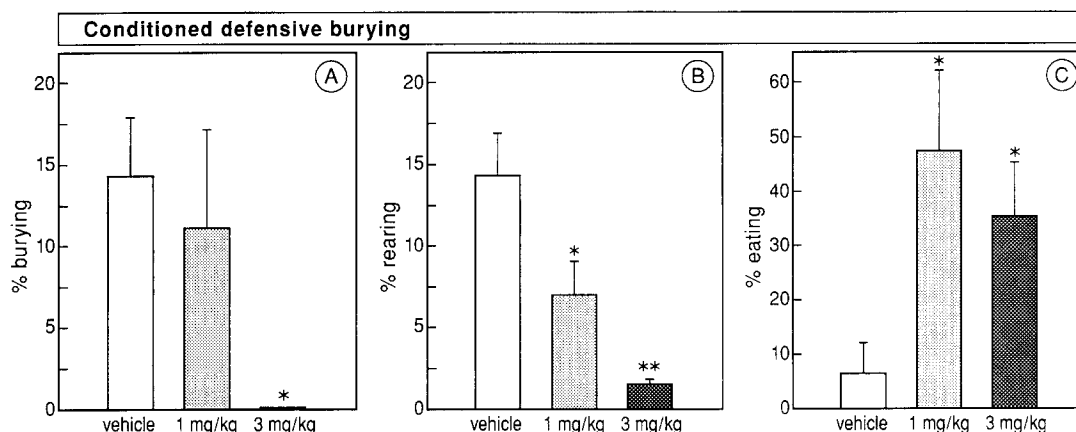
Fig. 2. Percent time spent on different types of behaviour during 10-min conditioned defensive burying test, 30 min after i.p. administered vehicle ($n = 7$) or R-(+)-8-OSO$_2$CF$_3$-PAT (1 mg/kg, $n = 6$; 3 mg/kg, $n = 6$). The different types of behaviour: defensive burying (A); rearing (B); eating (C). Data are expressed as means ± S.E.M. * $P \leq 0.05$; ** $P \leq 0.01$, significantly different from control.
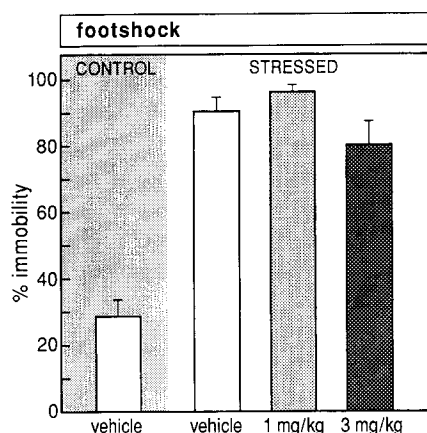

Fig. 3. Percent time spent immobile during exposure to the former footshock compartment, 30 min after i.p. administered vehicle (control, $n = 6$; stressed, $n = 8$) or R-(+)-8-OSO$_2$CF$_3$-PAT (1 mg/kg, $n = 7$; 3 mg/kg, $n = 7$). Data are expressed as means ± S.E.M. * $P \leq 0.05$, significantly different from control.

somatosensory cortex ($F(2,19) = 5.11$; $P \leq 0.05$), dorsal hippocampus ($F(2,26) = 3.99$; $P \leq 0.05$) and lateral septum ($F(2,25) = 4.91$; $P \leq 0.05$). In the case of the lateral septum, the maximum effect was achieved at a dose of 1 mg/kg ($P \leq 0.05$). The 5-HIAA/5-HT ratio of the paraventricular nucleus of the hypothalamus and the ventral median hypothalamus showed a trend towards a decrease, whereas the ratios in other brain regions were not altered (Table 1).

4. Discussion

The treatment with R-(+)-8-OSO$_2$CF$_3$-PAT, recently reported to be a chemically and biologically stable 5-HT$_{1A}$ receptor agonist (Sonesson et al., 1993, 1995), led to an increase in time spent in the open arms of the plus-maze and decreased defensive burying behaviour. Therefore, it is
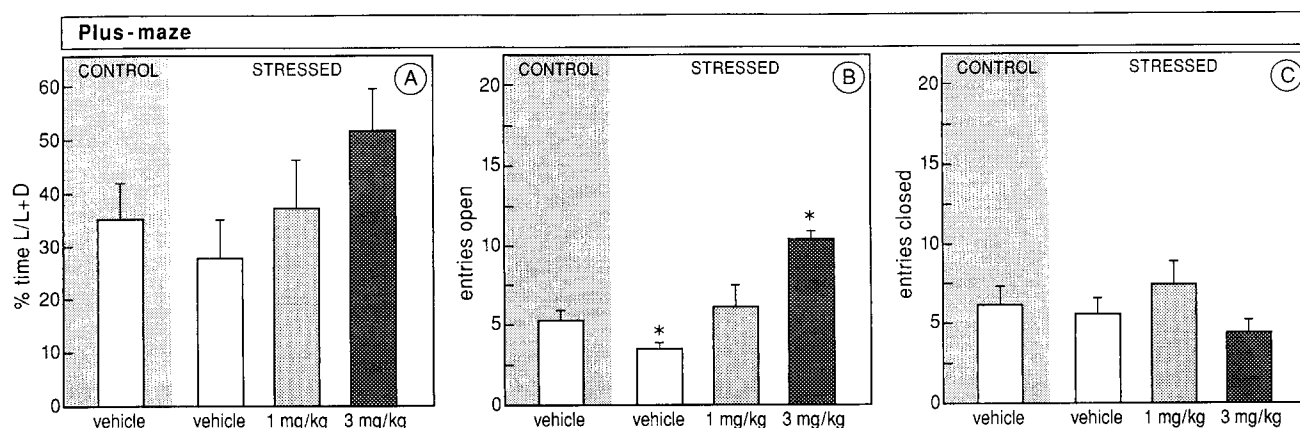

Fig. 4. The effect of R-(+)-8-OSO$_2$CF$_3$-PAT (1 and 3 mg/kg i.p.) on the (A) percent time spent in the open arms relative to cumulative time in all four arms; (B) number of open-arm entries; (C) number of closed-arm entries in rats given a 5-min test in the elevated plus-maze, directly after the rats' exposure to the conditioned emotional stressor. For further explanations see Fig. 2.

Table 1

The ratio of 5-HIAA/5-HT in median raphe nucleus (MRN), dorsal raphe nucleus (DRN), somatosensory cortex (SC), prefrontal cortex (PC), dorsal hippocampus (DH), lateral septum (LS), medial septum (MS), striatum, paraventricular nucleus of the hypothalamus (PVN), ventral median hypothalamus (VMH), central amygdala (CEA) and accumbens, 30 min after i.p. administration of saline ($n = 8$ or 9) or R-(+)-8-OSO$_2$CF$_3$-PAT (1 mg/kg, $n = 5$–9; 3 mg/kg, $n = 6$–9)

Brain area	Control	R-(+)-8-OSO$_2$CF$_3$-PAT (1 mg/kg)	R-(+)-8-OSO$_2$CF$_3$-PAT (3 mg/kg)
MRN	1.809 ± 0.182	2.260 ± 0.384	2.444 ± 0.504
DRN	1.155 ± 0.074	1.338 ± 0.127	0.979 ± 0.052
SC	1.701 ± 0.191	1.345 ± 0.122	1.009 ± 0.032 [*]
PC	1.834 ± 0.203	1.935 ± 0.233	1.386 ± 0.183
DH	1.487 ± 0.166	1.249 ± 0.064	1.061 ± 0.050 [*]
LS	1.101 ± 0.099	0.755 ± 0.091 [*]	0.784 ± 0.070 [*]
MS	1.081 ± 0.124	0.893 ± 0.137	0.977 ± 0.130
Striatum	1.188 ± 0.083	1.052 ± 0.124	1.124 ± 0.087
PVN	0.902 ± 0.086	0.916 ± 0.208	0.722 ± 0.117
VMH	1.341 ± 0.116	1.278 ± 0.114	1.051 ± 0.130
CEA	0.929 ± 0.128	0.828 ± 0.126	0.827 ± 0.091
Accumb	1.047 ± 0.060	1.417 ± 0.344	1.367 ± 0.245

Data are expressed as means ± S.E.M. [*] $P \leq 0.05$, significantly different from control.

suggested that this new compound possesses anxiolytic properties. The same dose range of this drug induced a reduction of the 5-HIAA/5-HT ratio in several parts of the limbic system.

Together, these behavioural animal models give a particularly strong indication of the anxiolytic properties of a drug. In the burying model the major index of anxiety is expressed as an active coping behaviour (burying; Korte and Bohus, 1990), whereas in the plus-maze test reduced exploration of the open arms is the major index of anxiety (Pellow et al., 1985; Treit et al., 1993). Decreases in defensive burying behaviour are evoked by classical benzodiazepine anxiolytics (De Boer et al., 1990; Treit and Fundytus, 1988) or 5-HT$_{1A}$ receptor agonists (e.g. ipsapirone, buspirone; Korte and Bohus, 1990; Treit and Fundytus, 1988) and are often interpreted as anxiolytic actions. In the burying test, R-(+)-8-OSO$_2$CF$_3$-PAT produced a dose-dependent decrease in burying behaviour and similar results were obtained for rearing activity. Interestingly, R-(+)-8-OSO$_2$CF$_3$-PAT at the doses applied induced hyperphagia during the conditioned defensive burying experiment. It is known that agonistic action at somatodendritic autoreceptors of e.g. 8-OH-DPAT, buspirone and gepirone reduces the synthesis and release of brain serotonin and thereby enhances food intake in freely feeding rats (Cooper, 1987, 1989; Dourish et al., 1985; Hutson et al., 1986, 1988a; Curzon, 1990). In addition, there is evidence that 5-HT levels in the hypothalamus mediate the regulation of food intake via 5-HT$_{1B}$ receptors. Direct infusion of 5-HT$_{1B}$ receptor agonists into the paraventricular nucleus induces hypophagia (Hutson et al., 1988b; Curzon, 1990; Macor et al., 1990). This suggests that lowering of the 5-HT level in the paraventricular nucleus may cause hyperphagia. However, this hypothesis cannot be supported or contradicted by our findings, since systemically administered R-(+)-8-OSO$_2$CF$_3$-PAT failed to sig-

nificantly reduce the 5-HIAA/5-HT ratio in the paraventricular nucleus of the hypothalamus.

Contradictory results have been reported for the effects of acutely administered partial or full 5-HT$_{1A}$ receptor agonists in the elevated plus-maze model (Handley and McBlane, 1993b). Some of these ligands were found to be anxiolytic, whereas other workers classed the same compounds as anxiogenic. Changes in experimental conditions may have dramatic effects, e.g., increasing the light intensity from 170 to 785 Lux causes inversion of the previously found anxiogenic effect (Handley and McBlane, 1993b). In our elevated plus-maze model, the anxious behaviour was enhanced by prior exposure to a conditioned stressor, i.e. re-exposure to a compartment associated with an inescapable, uncontrollable stressor (Korte et al., 1995). This was manifested as diminished exploration of the open arms of the plus-maze, as compared to the non-stressed controls. Acute treatment with 3 mg/kg of R-(+)-8-OSO$_2$CF$_3$-PAT not only prevented this effect but even resulted in enhanced exploration behaviour in the open arms, as compared to the non-stressed control group. The number of entries to enclosed arms of the non-stressed, stressed and treated groups remained unchanged.

Obviously, R-(+)-8-OSO$_2$CF$_3$-PAT could not block the shock-induced immobility in the inescapable footshock compartment at the doses tested. Footshock-induced freezing provides one way of examining the anxiolytic potential of drugs from a number of different classes. At the dose of 2.5 mg/kg, but not at the dose of 5.0 mg/kg, buspirone reduced footshock-induced freezing (Conti et al., 1990). Ipsapirone (12.5 mg/kg) reduced the conditioned immobility behaviour, not only in stressed, but also in non-stressed animals (Korte and Bohus, 1990). Therefore, in the latter case, it remained questionable whether ipsapirone has an anxiolytic action or whether it acts as stimulant on behavioural activity in general. So far, effects of other

(partial) 5-HT$_{1A}$ receptor agonists in this model have not been reported. Considering the anxiolytic efficacy of R-(+)-8-OSO$_2$CF$_3$-PAT in the other two anxiety models, it is possible that a different form of anxiety is generated in this model. The differential effect in the footshock compartment suggests that its anxiolytic effects may depend on the type of stimulus used to induce fear. Furthermore, it is postulated that different forms of experimental anxiety may be modulated in different ways by specific serotonin receptor subtypes (Treit et al., 1993).

So far, only scarce data have been presented on the relationship between (partial) 5-HT$_{1A}$ receptor agonists, 5-HT turnover in particular brain areas and stress. Saphier and Welch (1995) examined the effects of 8-OH-DPAT on neurochemical responses in various stress models. Conditioned fear-induced increases in the 5-HIAA/5-HT ratio in the prefrontal cortex were attenuated by 8-OH-DPAT; however, stress-induced excitatory output as a result of footshock could not be inhibited by 8-OH-DPAT. The potential anxiolytic and partial 5-HT$_{1A}$ receptor agonist, ipsapirone (5 mg/kg i.p.), reduces 5-HIAA/5-HT ratios [3] in various brain areas (brainstem, hypothalamus, striatum, hippocampus, anterior cortex and posterior cortex) by 40–50% (Hamon et al., 1988). Buspirone, another partial 5-HT$_{1A}$ receptor agonist, mimics the inhibitory activity of 5-HT to suppress neuronal activity in the dorsal raphe nuclei after systemic application (Vandermaelen et al., 1986) and produces a decrease in cortical 5-HIAA levels (40%), comparable to decreases evoked by the same dose of ipsapirone (Hamon et al., 1988). However, like R-(+)-8-OSO$_2$CF$_3$-PAT, but unlike ipsapirone, buspirone had no effect on striatal 5-HIAA and 5-HT levels (Cimino et al., 1983). The limbic, striatal and cortical, but not hippocampal, 5-HIAA/5-HT ratios were decreased by intra-DRN 8-OH-DPAT infusion (Hjorth and Magnusson, 1988). Liu et al. (1993, 1995) found a decreased 5-HIAA/5-HT ratio (up to 49%) in hippocampal rat brain homogenates after s.c. administration of 5-HT$_{1A}$ receptor agonists. The reference compound, 8-OH-DPAT, lowered 5-HT turnover by 39% at a dose of 1 μmol/kg. In the present study, administration of R-(+)-8-OSO$_2$CF$_3$-PAT induced a significant reduction of the 5-HIAA/5-HT ratio in the somatosensory cortex, dorsal hippocampus and lateral septum (41, 29 and 31%, respectively). The latter result can be compared with findings of Treit et al. (1993), who reported a decrease in defensive burying after septal and raphe lesions. Similarly, we observed abolition of defensive burying after administration of R-(+)-8-OSO$_2$CF$_3$-PAT. This suggests that the projection of the raphe to the septum might be particularly important. Trends towards a decrease of the 5-HIAA/5-HT ratio were found in paraventricular nucleus of the hypothalamus, the ventral me-

dian hypothalamus and central amygdala. Together, these findings suggest that serotonergic function after R-(+)-8-OSO$_2$CF$_3$-PAT administration is reduced in the limbic system in general. The limbic structures are predominantly innervated by the dorsal raphe nucleus and the median raphe nucleus (Azmitia and Segal, 1978). Therefore, it is assumed that the observed anxiolytic effects are mediated by both nuclei, and not exclusively by either of these brain areas. Direct stimulation of dorsal hippocampal 5-HT$_{1A}$ receptors may also produce anxiolytic effects, e.g., intradorsal hippocampal administration of 8-OH-DPAT increases exploration of open arms in the plus-maze (Guimaraes et al., 1994). Przegaliński et al. (1994) found anxiolytic-like effects of ipsapirone in the conflict drinking test after injection into the dorsal hippocampus.

In conclusion, the present results suggest that acute administration of the 5-HT$_{1A}$ receptor agonist, R-(+)-8-OSO$_2$CF$_3$-PAT, induces anxiolytic effects by stimulation of the presynaptic 5-HT$_{1A}$ somatodendritic receptors in the raphe nuclei, resulting in a decrease of 5-HT neurotransmission in somatosensory cortex, dorsal hippocampus and lateral septum. Therefore, it is assumed that these brain regions may play an important role in the behavioural expression of anxiety. However, it cannot be excluded that a direct agonistic action on postsynaptic 5-HT$_{1A}$ receptors in the dorsal hippocampus contributes to this anxiolytic effect.

Acknowledgements

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