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Involvement of somatodendritic 5-HT_{1A} receptors in Δ^9-tetrahydrocannabinol-induced hypothermia in the rat

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Abstract

Previously, it has been reported that modulating serotonergic neurones by use of selective serotonin reuptake inhibitors (SSRI) can alter the hypothermic response produced by Δ^9-tetrahydrocannabinol (Δ^9-THC). The aim of the present study was to investigate the effect that activation or antagonism of 5-hydroxytryptamine (5-HT_{1A}) receptors has on Δ^9-THC-induced hypothermia. Δ^9-THC (0.5, 2 and 5 mg/kg iv) decreased body temperature in a dose-related manner. Whilst having no significant effect on body temperature when administered 40 min prior to vehicle injection, the 5-HT_{1A} receptor antagonist *N*-[2-[4-(2-methoxyphenyl)-1-piperazinyl]ethyl]-*N*-(2-pyridinyl) cyclohexanecarboxamide trihydrochloride (WAY 100635; 1 mg/kg sc) significantly potentiated the hypothermia produced by 2 and 5 mg/kg Δ^9-THC. In order to investigate whether this effect was due to antagonism at somatodendritic autoreceptors in midbrain raphe nuclei, WAY 100635 or the 5-HT_{1A} agonist 8-hydroxy-(di-*n*-propylamino) tetralin (8-OH-DPAT) was microinjected into either the median raphe nuclei (MRN) or dorsal raphe nuclei (DRN) 40 min prior to Δ^9-THC injection. Following microinjection into the DRN, neither WAY 100635 (0.5 nmol/0.5 μl/10 s) nor 8-OH-DPAT (15.2 nmol/0.5 μl/10 s) had any significant effect on Δ^9-THC-induced hypothermia. However, WAY 100635 when microinjected into the MRN significantly potentiated Δ^9-THC-induced hypothermia, and 8-OH-DPAT microinjected into the MRN significantly inhibited Δ^9-THC-induced hypothermia. It is suggested from these studies that the potentiation of Δ^9-THC-induced hypothermia by WAY 100635 when administered peripherally is mainly due to antagonism at somatodendritic 5-HT_{1A} autoreceptors in the MRN.

Keywords: Δ^9-THC; 5-HT; 5-HT_{1A} receptor; Hypothermia

1. Introduction

Δ^9-tetrahydrocannabinol (Δ^9-THC), the major psychoactive constituent of cannabis, produces a significant dose-dependent hypothermic response, which occurs via a central mechanism (Fitton and Pertwee, 1982). This effect occurs irrespective of the route of administration or the animal species used (Hardman et al., 1971; Holtzman et al., 1969) and can be blocked by the cannabinoid CB_1 receptor antagonist *N*-piperidino-5-(4-chlorphenyl)-1-(2,4-dichlorophenyl)-4-methyl-3-pyrazole-carboxamide hydrochloride (SR 141716A; Compton et al., 1996; Malone and Taylor, 1998b), indicating that the effect is mediated by CB_1 receptors. Along with antinociception and decreased locomotor activity, the cannabinoid-induced hypothermia provides a good quantitative measure of brain cannabinoid activity and is particularly useful when evaluating the cannabimimetic activity of cannabinoid analogues (Compton et al., 1992) and recently discovered endogenous cannabinoids such as anandamide (Lichtman et al., 1996).

It has long been recognised that 5-hydroxytryptamine (5-HT; serotonin) is involved in the central regulation of body temperature (Feldberg and Lotti, 1967; Feldberg and Myers, 1964). Also, several lines of evidence support the view that 5-HT may be involved in the cannabinoid-induced hypothermic response. For example, pretreatment with the selective serotonin reuptake inhibitor (SSRI) clomipramine has been shown to modify the hypothermia induced by Δ^9-THC and alter Δ^9-THC-induced alterations in concentrations of brain 5-hydroxyindoleacetic acid (5-HIAA) in the rat (Fennessy and Taylor, 1978). More recently, we published a study that showed how CB_1 receptor-mediated hypothermia could be augmented or antagonised by administration of the SSRI

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fluoxetine (Malone and Taylor, 1998b). The direction of the observed modulation was dependent on the time of administration of fluoxetine relative to that of Δ^9-THC. Pretreatment with fluoxetine increases extracellular 5-HT as a result of inhibition of 5-HT reuptake. Increased extracellular 5-HT can activate autoreceptors, which may decrease serotonergic activity, thereby reducing the Δ^9-THC-induced hypothermia. Conversely, when fluoxetine is administered after Δ^9-THC, the reuptake block is thought to potentiate the already activated serotonergic system, hence potentiating the Δ^9-THC-induced hypothermia. From these results, we hypothesised that the amount of 5-HT in the synaptic cleft influenced the postjunctional effects observed by altering the activity of the 5-HT neurones. Furthermore, we speculated that CB_1 receptor activation by Δ^9-THC results in an increase in extracellular 5-HT in order to produce hypothermia.

In order to further investigate this hypothesis, the effects of pretreatment with the selective 5-HT_{1A} antagonist *N*-[2-[4-(2-methoxyphenyl)-1-piperazinyl]ethyl]-*N*-(2-pyridinyl) cyclohexanecarboxamide trihydrochloride (WAY 100635) and the 5-HT_{1A} agonist 8-hydroxy-(di-*n*-propylamino) tetralin (8-OH-DPAT) on the Δ^9-THC-induced hypothermic response has been examined. Some of these results have been presented at the Australasian Society of Clinical and Experimental Pharmacologists and Toxicologists (ASCEPT) 1998 meeting (Malone and Taylor, 1998a).

2. Methods

2.1. Animals

Female albino Glaxo–Wistar rats weighing between 200 and 250 g were used. Prior to experimentation, they were housed in group cages and kept at 22°C with a 12 h light–dark cycle. Food and water were available ad libitum.

2.2. Surgery

For intravenous (iv) administration of drugs, rats were implanted with permanent polyethylene (PE 50) catheters into the external jugular vein under methohexitone sodium (18 mg/kg ip)/amylobarbitone sodium (30 mg/kg ip) anaesthesia. For rats receiving microinjection into the dorsal raphe nucleus (DRN) or the median raphe nucleus (MRN), the animals were then placed in a stereotaxic frame. The skull was exposed and a hole (2 mm i.d.) was drilled for the implantation of a stainless-steel 23 G guide cannula that was positioned above the DRN and MRN (coordinates: L 0 mm, R +1.2 mm). Animals were implanted with the guide cannula (V −4.2 mm), which was held in place using dental cement anchored to two stainless-steel screws implanted into the skull. A stainless-steel 30 G needle, cut to size, was inserted into the cannula as an obturator. The coordinates were chosen according to the stereotaxic atlas of Paxinos and Watson (1986). Body temperature was maintained at 37°C with a heating pad used throughout the course of the operation. To minimise the incidence of postoperative infection, animals were injected with ticarcillin (15 mg/kg ip) immediately following the operation and daily for 5 days prior to the experiment. During the recovery period, animals were kept in individual cages, and food and water were available ad libitum.

2.3. Experimental procedure

For each treatment group, 5–13 rats were used. The animals were gently restrained during the microinjection. The obturators were removed from the guide cannulae and a stainless-steel cannula connected by polyethylene tubing to a CMA/100 microinjection pump (Carnegie Medicin, Stockholm, Sweden) was inserted via the guide cannula (coordinates: DRN: V − 6.2 mm; MRN: V − 8.5 mm). WAY 100635 (0.5 nmol/0.5 μl/10 s), 8-OH-DPAT (15.2 nmol/0.5 μl/10 s) or saline (0.5 μl/10 s) was microinjected into either the DRN or the MRN. The dose of WAY 100635 (0.5 nmol/0.5 μl) used in this study was chosen based on a previous study, which showed that WAY 100635 (0.47 nmol/0.5 μl) microinjected into the DRN selectively antagonised the effects of 8-OH-DPAT (Carli et al., 1998). The dose of 8-OH-DPAT (15.2 nmol/0.5 μl) was chosen based on a previous study, which showed that this dose produced a complete inhibition of the discharge of neurones from the DRN (Jolas et al., 1995). The cannula was left in place for 3 min after microinjection. After resealing the guide cannula with a stainless-steel stylet, the animals were returned to their home cage. The microinjection of WAY 100635, 8-OH-DPAT or saline was followed 40 min later by the intravenous administration of Δ^9-THC vehicle or 2 or 5 mg/kg Δ^9-THC. Body temperature and behavioural studies were all undertaken in the afternoon. Body temperature was recorded by means of a thermister probe inserted 6 cm into the colon. Core body temperature was recorded before WAY 100635, 8-OH-DPAT or saline pretreatment, immediately before Δ^9-THC or vehicle and at 30, 60, 90, 120, 150 and 1200 min after Δ^9-THC or vehicle administration.

At the end of experiments involving central microinjections, each rat was killed with an overdose of pentobarbitone. A stainless-steel cannula was inserted via the guide cannula as above and black dye (0.5 μl/10 s) was microinjected into either the DRN or the MRN. The cannula was left in place for 3 min after microinjection. The brain was quickly removed and frozen, and coronal sections were cut to verify the correct position of the microinjection needle. Only results obtained from rats with correctly positioned injection cannulae are presented.

2.4. Drugs

Δ^9-THC (SIGMA) was incorporated into the triglyceride/phospholipid emulsion Intralipid as described previously (Malone and Taylor, 1998b). A concentration of 3 mg/ml

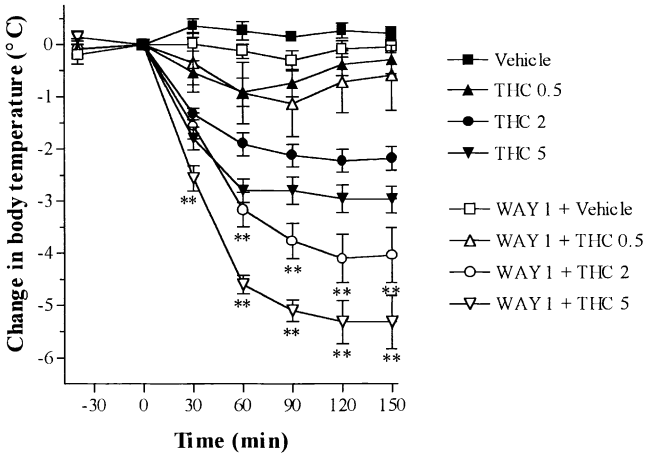

Fig. 1. The effect of pretreatment (at $t=-40$ min) with WAY 100635 1 mg/kg sc (WAY 1) on hypothermia produced by intravenous Δ^9-THC (THC; filled symbols represent Δ^9-THC or vehicle alone while open symbols represent WAY 100635 + Δ^9-THC or vehicle). Data are expressed as a change in body temperature from that recorded immediately prior to Δ^9-THC or vehicle. For 2 mg/kg Δ^9-THC, WAY 100635 significantly potentiated body temperature at 60 [ANOVA, $F(3,27)=53.49$, $P<.01$], 90 [ANOVA, $F(3,27)=59.28$, $P<.01$], 120 [ANOVA, $F(3,27)=53.82$, $P<.01$] and 150 min [ANOVA, $F(3,27)=50.35$, $P<.01$]. For 5 mg/kg Δ^9-THC, WAY 100635 potentiated body temperature at 30 [ANOVA, $F(3,23)=44.64$, $P<.01$], 60 [ANOVA, $F(3,23)=117.55$, $P<.01$], 90 [ANOVA, $F(3,23)=115.12$, $P<.01$], 120 [ANOVA, $F(3,23)=97.99$, $P<.01$] and 150 min [ANOVA, $F(3,23)=86.49$, $P<.01$]. ** $P<.01$, when compared with Δ^9-THC alone.

Δ^9-THC in Intralipid was used for 2 and 5 mg/kg doses and 0.5 mg/ml Δ^9-THC in Intralipid was used for 0.5 mg/kg doses. WAY 100635 was dissolved in water for injection, and when injected peripherally, doses of 1 mg/kg sc were used. When injected centrally, WAY 100635 and 8-OH-DPAT were dissolved in water for injection.

Other drugs used were amylobarbitone sodium (Ely Lilly), methohexitone sodium (Eli Lilly), Intralipid (Baxter), WAY 100635 (Wyeth) and 8-OH-DPAT (Tocris).

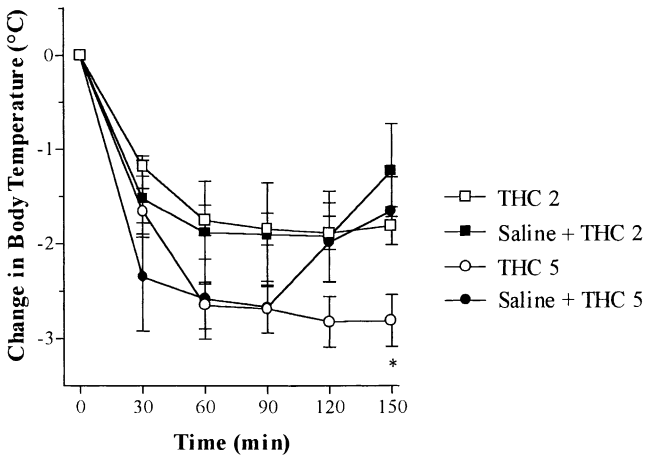

Fig. 2. Effect of Δ^9-THC (THC) on body temperature of rats with implanted microinjection cannulae and microinjected with saline (0.5 μl/10 s; administered at $t=-40$ min) compared with the body temperature of rats with no microinjection cannulae implanted (* $P<.05$, $n=6-11$). Data are expressed as a change in body temperature from that recorded immediately prior to Δ^9-THC or vehicle (at $t=0$). ANOVA main effect, saline + 2 mg/kg Δ^9-THC: $F(4,20)=0.39$, $P>.05$; ANOVA main effect, saline + 5 mg/kg Δ^9-THC: $F(4,20)=0.14$, $P>.05$. For 5 mg/kg Δ^9-THC, there was a significant reduction in hypothermia produced by rats injected with saline + Δ^9-THC compared with Δ^9-THC alone (t test, $P<.05$).

2.5. Statistical analysis of data

When comparing values following pretreatment plus Δ^9-THC or vehicle, a one-way analysis of variance

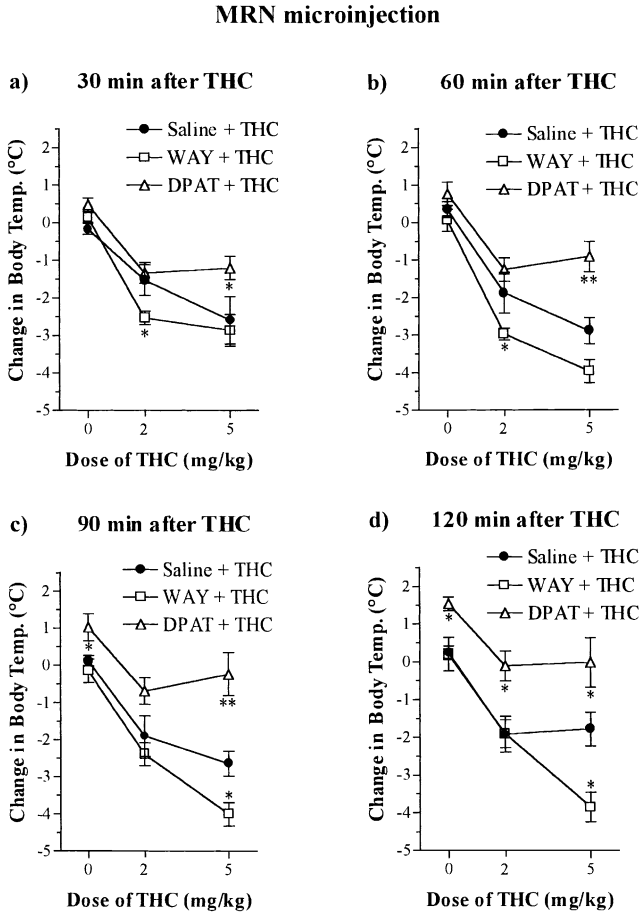

Fig. 3. Effect of WAY 100635 (WAY; 0.5 nmol/0.5 μl/10 s), 8-OH-DPAT (DPAT; 15.2 nmol/0.5 μl/10 s) or saline (0.5 μl/10 s) microinjected into the MRN 40 min prior to Δ^9-THC (THC) injection on body temperature. Data shown are at (A) 30 min [8-OH-DPAT vs. THC vehicle, ANOVA $F(2,12)=4.55$, $P<.05$; WAY + 2mg/kg THC vs. 2 mg/kg THC, ANOVA $F(2,18)=6.86$, $P<.05$; DPAT + 5 mg/kg THC vs. 5 mg/kg THC, ANOVA $F(2,14)=4.077$, $P<.05$]; (B) 60 min [WAY + 2mg/kg THC vs. 2 mg/kg THC, ANOVA $F(2,18)=9.14$, $P<.05$; DPAT + 5 mg/kg THC vs. 5 mg/kg THC, ANOVA $F(2,14)=19.74$, $P<.01$]; (C) 90 min [8-OH-DPAT vs. THC vehicle, ANOVA $F(2,12)=4.42$, $P<.05$; DPAT + 5 mg/kg THC vs. 5 mg/kg THC, ANOVA $F(2,14)=20.12$, $P<.01$; WAY + 5mg/kg THC vs. 5 mg/kg THC, ANOVA $F(2,14)=20.12$, $P<.05$] and (D) 120 min [8-OH-DPAT vs. THC vehicle, ANOVA $F(2,12)=6.76$, $P<.05$; DPAT + 2 mg/kg THC vs. 2 mg/kg THC, ANOVA $F(2,18)=6.43$, $P<.05$; DPAT + 5 mg/kg THC vs. 5 mg/kg THC, ANOVA $F(2,14)=14.51$, $P<.05$; WAY + 5 mg/kg THC vs. 5 mg/kg THC, ANOVA $F(2,14)=14.51$, $P<.05$] after Δ^9-THC injection. * $P<.05$, ** $P<.01$ when compared with Δ^9-THC alone (closed symbols). Data are expressed as a change in body temperature from that recorded immediately prior to Δ^9-THC or vehicle (at $t=0$).

(ANOVA) was used. When values were found to be significant ($P<.05$), the Student–Newman–Keuls Multi-comparison Test was used to determine the treatment groups that were different and the level of significance ($P<.05$ or .01). When comparing the effect of Δ^9-THC in animals with microinjection cannulae implanted and saline injected 40 min before Δ^9-THC to the effect of Δ^9-THC in animals without microinjection cannulae, a Student's t test was used.

3. Results

Administration of Δ^9-THC resulted in a dose-dependent decrease in body temperature following intravenous administration. The maximum decrease following the 0.5 mg/kg dose was 0.8 ± 0.2°C and occurred 60 min after Δ^9-THC administration. Maximum decreases of 2.0 ± 0.2°C and 2.9 ± 0.3°C occurred after doses of 2 and 5 mg/kg Δ^9-THC, respectively, and occurred at 120 min after Δ^9-THC administration (Fig. 1). The hypothermia was significant when compared with vehicle alone during the first 90 min after the 0.5 mg/kg dose of Δ^9-THC. Following the 2 and 5 mg/kg doses, the hypothermic response was significantly different from vehicle alone throughout the duration of the experiment.

WAY 100635 (1 mg/kg sc) produced no significant effect on body temperature when administered alone (Fig. 1). WAY 100635 (1 mg/kg sc) significantly potentiated the hypothermia produced by the 2 and 5 mg/kg doses of Δ^9-THC but had no significant effect on the hypothermia observed following 0.5 mg/kg Δ^9-THC (Fig. 1). Maximum decreases of 4.1 ± 0.5°C and 5.3 ± 0.4°C occurred after WAY 100635 + 2 mg/kg and WAY 100635 + 5 mg/kg Δ^9-THC, respectively, at 120 min after Δ^9-THC administration (Fig. 1). Thus, WAY 100635 was microinjected into the DRN and MRN prior to 2 and 5 mg/kg doses of Δ^9-THC only (see below).

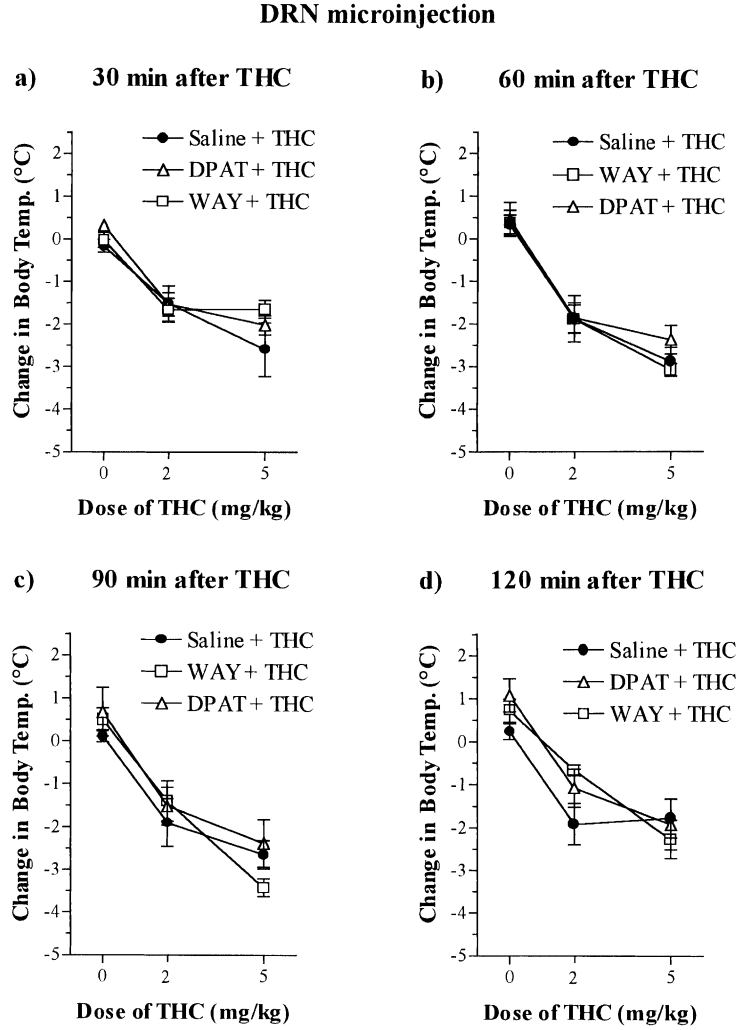

Fig. 4. Effect of WAY 100635 (WAY; 0.5 nmol/0.5 μl/10 s), 8-OH-DPAT (DPAT; 15.2 nmol/0.5 μl/10 s) or saline (0.5 μl/10 s) microinjection into the DRN 40 min prior to Δ^9-THC (THC) injection on body temperature. Data shown are at (a) 30, (b) 60, (c) 90 and (d) 120 min after Δ^9-THC injection. Data are expressed as a change in temperature from that recorded immediately prior to Δ^9-THC or vehicle (at $t=0$).

Saline microinjection (0.5 μl/10 s) into the MRN had no significant effect on Δ^9-THC-induced hypothermia during the first 90 min following Δ^9-THC injection (Fig. 2). Saline microinjection significantly reduced the hypothermia produced by 5 mg/kg Δ^9-THC at 150 min after Δ^9-THC injection but had no significant effect on 2 mg/kg Δ^9-THC. As saline microinjection had no significant effect on Δ^9-THC-induced hypothermia for the majority of the experiment, and in order to reduce the number of rats used, all microinjection studies involving WAY 100635 or 8-OH-DPAT pretreatments + Δ^9-THC were compared with MRN saline + Δ^9-THC values obtained.

WAY 100635 (0.5 nmol/0.5 μl/10 s) microinjected into the DRN or MRN had no significant effect on body temperature (data not shown). WAY 100635 (0.5 nmol/0.5 μl/10 s) microinjected into the MRN significantly potentiated the hypothermia produced by the 2 and 5 mg/kg doses of Δ^9-THC. This effect occurred in the first hour after 2 mg/kg Δ^9-THC but remained significantly different from Δ^9-THC alone following 5 mg/kg Δ^9-THC for 120 min after Δ^9-THC administration (Fig. 3). WAY 100635 (0.5 nmol/0.5 μl/10 s) microinjected into the DRN did not significantly potentiate the hypothermia produced by the 2 mg/kg dose of Δ^9-THC (Fig. 4). Microinjection of WAY 100635 into the DRN had no significant effect on hypothermia produced by the 5 mg/kg dose of Δ^9-THC.

8-OH-DPAT (15.2 nmol/0.5 μl/10 s) significantly increased body temperature at 130 and 160 min after microinjection into the MRN (90 and 120 min after vehicle administration; Fig. 3). 8-OH-DPAT (15.2 nmol/0.5 μl/10 s) microinjected into the MRN significantly attenuated the hypothermia produced by 2 mg/kg Δ^9-THC 120 min after Δ^9-THC injection (Fig. 3). Microinjection of 8-OH-DPAT into the MRN significantly attenuated the hypothermia produced by 5 mg/kg Δ^9-THC from 30 to 120 min after Δ^9-THC injection.

In contrast, 8-OH-DPAT (15.2 nmol/0.5 μl/10 s) had no significant effect on body temperature following microinjection into the DRN (Fig. 4). Also, no significant change in the hypothermic effect of 2 and 5 mg/kg Δ^9-THC was observed when 8-OH-DPAT (15.2 nmol/0.5 μl/10 s) was microinjected into the DRN 40 min prior to Δ^9-THC.

The body temperature change following either subcutaneous (sc) or microinjection of WAY 100635 + Δ^9-THC, 8-OH-DPAT + Δ^9-THC or saline + Δ^9-THC had returned to control levels when measured 1200 min after Δ^9-THC administration (data not shown).

4. Discussion

The present study adds further support to earlier investigations demonstrating that altering the activity of serotonergic neurones in rats (Fennessy and Taylor, 1978; Malone and Taylor, 1998b) and mice (Davies and Graham, 1980) can modify Δ^9-THC-induced hypothermia.

WAY 100635 has been shown to be a potent and selective antagonist at 5-HT_{1A} receptors (Forster et al., 1995). WAY 100635 alone, when administered either centrally or peripherally, had no significant effect on body temperature. This is in agreement with previous studies, which have reported that whilst WAY 100635 can antagonise the hypothermia produced by 5-HT_{1A} agonists (Fletcher et al., 1996; Forster et al., 1995; Patel and Hutson, 1996), WAY 100635 alone produces no significant effects on body temperature (Forster et al., 1995).

WAY 100635 (1 mg/kg sc) in the present study significantly potentiated the hypothermic response induced by the 2 and 5 mg/kg doses of Δ^9-THC. Brain regions containing 5-HT_{1A} receptors include the hippocampus, midbrain raphe nuclei, amygdala, septum and various regions within the hypothalamus (Pompeiano et al., 1992). Thus, WAY 100635 is presumably acting in one or more of these regions in order to potentiate Δ^9-THC-induced hypothermia. Previous studies have shown that in the rat, 5-HT_{1A} receptor agonist-induced hypothermia is mediated by postsynaptic 5-HT_{1A} receptors (Bagdy and To, 1997; Bill et al., 1991). Also, it has been speculated that CB_1 receptor activation by Δ^9-THC results in an increase in extracellular 5-HT in order to produce hypothermia (Malone and Taylor, 1998b). Hence, if WAY 100635 when injected peripherally was predominantly blocking postsynaptic 5-HT_{1A} receptors, then an antagonism of Δ^9-THC-induced hypothermia would be expected as opposed to the potentiation observed in the present study. Thus, in the present study, it is assumed that WAY 100635 is blocking presynaptic 5-HT_{1A} receptors in order to potentiate Δ^9-THC-induced hypothermia.

Antagonism at somatodendritic 5-HT_{1A} receptors in midbrain raphe regions has been shown to potentiate the effects of drugs that increase extracellular 5-HT levels such as SSRIs (Gartside et al., 1995; Invernizzi et al., 1997; Sharp et al., 1997). Specific regions within the hypothalamus are thought to play an important role in the regulation of body temperature (Cox and Lee, 1981). For example, neurones responsive to thermal stimulation of the skin exist in the preoptic region and the anterior and posterior regions of the hypothalamus (Bruck and Hinckel, 1982). As the hypothalamus receives input from serotonergic neurones with cell bodies in the DRN and the MRN (Azmitia and Segal, 1978; Moore et al., 1978), the blockade of somatodendritic 5-HT_{1A} autoreceptors in either or both of these regions by WAY 100635 may be responsible for potentiating the Δ^9-THC-induced hypothermia.

In order to investigate this further, WAY 100635 was microinjected into either the DRN or the MRN prior to Δ^9-THC administration. Microinjection of WAY 100635 into the MRN (but not the DRN) significantly potentiated the Δ^9-THC-induced hypothermia. This may suggest that 5-HT neurones projecting from the MRN to the hypothalamus play a more significant role in the mechanism of Δ^9-THC in producing hypothermia than those from the DRN. In support of this observation is the work of Van de Kar and Lorens

(1979) who showed that electrolytic lesions in the MRN but not the DRN significantly decreased the 5-HT content of the anterior hypothalamus (Van de Kar and Lorens, 1979).

In order to further confirm the involvement of somatodendritic 5-HT_{1A} receptors in modulating Δ^9-THC-induced hypothermic response, the 5-HT_{1A} receptor agonist 8-OH-DPAT was microinjected into the DRN or the MRN prior to Δ^9-THC. Microinjection of 8-OH-DPAT into the MRN significantly reduced hypothermia produced by Δ^9-THC, whereas microinjection of 8-OH-DPAT into the DRN had no significant effect. In addition, microinjection of 8-OH-DPAT into the MRN produced a significant increase in body temperature when compared with saline controls, whereas microinjection of 8-OH-DPAT into the DRN had no significant effect on body temperature. These results support the suggestion that the MRN is the region predominantly associated with modulating Δ^9-THC-induced hypothermia as opposed to the DRN.

Previous studies have reported that peripherally administered 8-OH-DPAT produces a decrease in body temperature (Bagdy and To, 1997; Bill et al., 1991; Millan et al., 1993; Wozniak et al., 1988) rather than an increase as observed in the present study following central administration. However, it is generally thought that, in rats, the hypothermia produced by peripheral 8-OH-DPAT is due to postsynaptic 5-HT_{1A} receptor activation (Bagdy and To, 1997). Thus, it is not surprising that in the present study, activation of presynaptic 5-HT_{1A} receptors by 8-OH-DPAT produced an increase in body temperature, since, as a consequence of a decrease in serotonergic neuronal firing, less extracellular 5-HT would be available to activate postsynaptic 5-HT_{1A} receptors.

As 8-OH-DPAT reduced and WAY 100635 potentiated Δ^9-THC-induced hypothermia, it is suggested that somatodendritic 5-HT_{1A} receptor activation attenuates, whereas 5-HT_{1A} receptor antagonism in the MRN potentiates Δ^9-THC-induced hypothermia. Such results are in agreement with previous work in our laboratory, from which the hypothesis was made that an increase in serotonergic activity results in a potentiation of Δ^9-THC-induced hypothermia, whereas a decrease in serotonergic activity decreases Δ^9-THC-induced hypothermia (Malone and Taylor, 1998b).

Autoradiographic tracing studies have shown that serotonergic neurones project from the MRN to virtually all hypothalamic areas (Azmitia and Segal, 1978; Moore et al., 1978). Thus, it appears likely that the effect of WAY 100635 and 8-OH-DPAT in modulating the effect of Δ^9-THC on body temperature is due to modulation of serotonergic neurones projecting from the MRN to the hypothalamus. However, the involvement of other serotonergic neurones projecting from the MRN cannot be ruled out.

Following histological verification of the position of the microinjection cannula, it was revealed that in animals whose microinjection cannulae lay outside the MRN, no significant change in Δ^9-THC-induced hypothermia was observed following microinjection of either WAY 100635 or 8-OH-DPAT (data not shown). Furthermore, there were significant differences in effect observed following WAY 100635 and 8-OH-DPAT microinjection into either the DRN or MRN. As the DRN and MRN are separated by approximately 1 mm (Paxinos and Watson, 1986), if significant diffusion occurred, the effects of microinjection into the DRN would not have been significantly different from microinjection into the MRN. Thus, it is suggested that the effects on Δ^9-THC-induced hypothermia produced by pretreatment with WAY 100635 or 8-OH-DPAT microinjected into the MRN are due to a specific effect on 5-HT_{1A} receptors within the MRN and not from diffusion into other brain regions away from the site of injection.

5. Conclusion

The results of the present study show that potentiation of Δ^9-THC-induced hypothermia by WAY 100635 when administered peripherally is mainly due to antagonism at somatodendritic 5-HT_{1A} autoreceptors in the MRN. Also, it is suggested that the ability of Δ^9-THC to produce hypothermia is dependent upon the normal firing rate of serotonergic neurones originating from the MRN.

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