

Serotonin 5-HT₄ receptors in the nucleus accumbens are specifically involved in the appetite suppressant and not locomotor stimulant effects of MDMA ('ecstasy')

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Abstract

Rationale 3,4-Methylenedioxymethamphetamine (MDMA) abuse is a substantial problem in young adults. Due to a high focus on body image in this population, two main factors that may encourage MDMA use are the appetite suppressant and locomotor stimulant effects of this drug. The nucleus accumbens (NAc) is a brain region associated with the regulation of motivated and locomotor behaviours, and recent evidence suggests that NAc 5-HT₄ receptors are likely to be involved in the appetite suppressant effect of MDMA. It has not yet been shown whether 5-HT₄ receptors of the NAc are involved in the locomotor stimulant effects of MDMA, which may also contribute to a reduction in food intake.

Objectives This study aimed to investigate the effect of local antagonism of serotonin 5-HT₄ receptors in the NAc in the appetite suppressant and locomotor stimulant effects of MDMA.

Methods Male hooded Wistar rats underwent surgery for the implantation of bilateral NAc microinjection cannulae under isoflurane anaesthesia. Following 5–7 days of recovery, the rats received bilateral microinjections of the 5-HT₄ antagonist RS39604 into the NAc immediately prior to either saline or MDMA administration. Food intake, water intake, body weight and locomotor activity were measured.

Results RS39604 significantly increased food intake and increased weight loss in MDMA-treated but not saline-treated rats. Measures of MDMA-induced water intake or locomotor activity were not altered by antagonist administration.

Conclusions These results demonstrate that 5-HT₄ receptors in the NAc specifically regulate the appetite suppressant effects of MDMA but not MDMA-induced water intake or locomotor activity.

Keywords Ecstasy · Nucleus accumbens · Appetite · Locomotor · Weight loss

Abbreviations

5-HT	Serotonin
MDMA	3,4 Methylenedioxymethamphetamine
NAc	Nucleus accumbens
ip	Intraperitoneal

Introduction

3,4-Methylenedioxymethamphetamine (MDMA or 'ecstasy') use is estimated to affect nine million people in the global population aged 15–64, with considerably high prevalence rates among the 20–29-year-old age group (United Nations Office on Drugs and Crime UNDOC 2008). The increased use of this drug, especially among 20–29 year olds, indicates a need for greater understanding of its effects and the factors motivating its use, as well as a need to develop prevention messages and treatment plans targeted at young adults (Degenhardt et al. 2009). Although MDMA is typically thought to appeal to users due to promoting euphoric and motor stimulant effects, the ages associated with recreational drug use (adolescence and early adulthood) are also associated

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with an increased focus on body image (Stock et al. 2002). Indeed, MDMA users have been found to have higher Eating Disorder Inventory scores than controls and be more likely than controls to agree with statements such as “ecstasy helps you lose weight” (Curran and Robjant 2006). Additionally, unhealthy weight control behaviours such as vomiting and laxative use have been demonstrated to be positively associated with past year MDMA use (Cance et al. 2005). Thus, two possible motivating factors for MDMA use are the focus of this study: the appetite suppressant effects of MDMA and the stimulant effects of MDMA that promote energy expenditure.

MDMA use results in alterations to monoaminergic signalling, where increases in the intracellular levels of the monoamines dopamine and serotonin have been measured in the nucleus accumbens (NAc) following MDMA administration (White et al. 1996). The NAc is a brain region that plays an important role in the regulation of feeding behaviour and motivation to consume food (Stratford and Kelley 1997; Jean et al. 2007) and is also well described for regulating locomotor activity through monoaminergic mechanisms (Kelly et al. 1975; Koob et al. 1978).

A role for serotonin in the regulation of food intake has been demonstrated by the suppression of appetite following treatments that increase extracellular serotonin levels, such as fenfluramine (Lucas et al. 1998; Vickers et al. 1999) and MDMA (Frith et al. 1987; Rochester and Kirchner 1999). Although the basic relationship between serotonin and food intake is well established (Blundell 1984), the involvement of specific serotonin receptors in appetite regulation is still being investigated. Recently, the 5-HT₄ receptor has been implicated in appetite regulation as stress-induced reductions in food intake were attenuated in 5-HT₄ receptor knockout mice (Compan et al. 2004).

There are an abundance of 5-HT₄ receptors in the NAc (Waeber et al. 1994), and the local administration of a 5-HT₄ receptor antagonist into the NAc produced an increase in food intake compared to controls in fed but not food-deprived mice (Jean et al. 2007). While this study by Jean et al. (2007) did not directly measure antagonism of 5-HT₄ receptors in the NAc on MDMA-induced appetite suppression, they demonstrated that MDMA-induced reductions in food intake were attenuated in 5-HT₄ receptor knockout mice, deprived of food for 18 h. Of interest to our study, food-deprived mice treated with MDMA showed increases in cocaine- and amphetamine-regulated transcript (CART) in the NAc of wild-type but not 5-HT₄ knockout mice, suggesting that the anorectic effects of MDMA were mediated by 5-HT₄ receptors in the NAc (Jean et al. 2007). Here, we extend the study by Jean et al. (2007) to investigating the effect of direct antagonism of 5-HT₄ receptors in the NAc on MDMA-induced appetite suppression in rats with free access to food and water.

In addition to appetite suppression, the consumption of MDMA produces stimulant effects to increase activity and movement in humans, effects that are exacerbated in environments such as dance clubs, where the rhythmic music encourages dancing for prolonged periods (Parrott 2001). These observations in humans are supported by a great number of animal studies demonstrating increased locomotor activity following MDMA administration (e.g. Gold et al. 1988; Spanos and Yamamoto 1989). It is possible that the perceptions of increased activity and weight loss induced by MDMA encourage repeated use and provide incentives to continue use of this drug.

Previous research suggests that 5-HT plays an important role in MDMA-induced locomotor activity (Callaway et al. 1991; Callaway et al. 1990); however, the specific receptors underlying serotonergic regulation of MDMA-induced locomotor activity have remained relatively unexplored. The high concentration of 5-HT₄ receptors in limbic structures, in particular the NAc, has led to suggestions that this receptor subtype is involved in motor behaviour (McMahon and Cunningham 1999; Reavill et al. 1998). In line with this, antagonism of 5-HT₄ receptors in the NAc has been shown to attenuate cocaine-induced locomotor activity (McMahon and Cunningham 1999).

In the present study, we investigated the effect of locally injecting RS39604, a high-affinity selective 5-HT₄ receptor antagonist (Hegde et al. 1995) into the NAc, on the appetite-suppressant effects of MDMA in rats with unrestricted access to food and water and on MDMA-induced motor stimulant behaviour. Water intake was also observed in order to determine the role of 5-HT₄ receptors in the effects of MDMA on consumptive behaviours in general.

Methods

Subjects

A total of 46 experimentally naïve male hooded Wistar rats aged between 2 and 3 months and weighing 200–300 g at the commencement of experimental proceedings were obtained from Adelaide University, Australia. The rats were housed individually in plastic tubs (60×37×18 cm) lined with wood shavings. Room temperature was maintained at 21°C on a 12-h reverse light–dark cycle with lights on at 20:00 hours. Food and water were available ad libitum in the home cages of the rats throughout the duration of the experiments but not during drug administration and test procedures. During testing procedures, they were provided with food and water as detailed below. All experiments were approved by the Macquarie University Animal Ethics Committee and complied with the Australian Code of Practice for the Care and Use of Animals for

Scientific Purposes (National Health and Medical Research Council 2004).

Experiment 1: MDMA dose-response curve for appetite suppression

To determine the dose of MDMA required to produce appetite suppressant effects, a total of 10 rats received four counterbalanced intraperitoneal (ip) injections of MDMA (2, 5 and 10 mg/kg) and vehicle (0.9% saline), following a latin square dosing regime. Drug administration occurred at 08:00 hours, the commencement of the rats' dark cycle, as the majority of the daily food consumption of rats occurs in the nocturnal phase (Cooper and Al-Naser 2006). Following drug administration, rats were returned to their home cages, where they were provided with 100 g of standard rat chow. At 1 and 3 h following drug administration, all remaining food was collected (including spilt food) and weighed. The rats were then immediately supplied with a further 100 g. This procedure was repeated until rats had received all four treatments, separated by at least 72 h.

Experiment 2: Food intake, water intake and body weight of rats administered RS39604

A total of 24 rats were used for this experiment. Rats were randomly assigned to receive ip injections of either MDMA (10 mg/kg, group 1: $n=12$) or saline (0.9%, group 2: $n=12$). Body weight and weight of water bottles were recorded immediately prior to drug administration. Drug administration commenced at 08:00 hours. Each group received bilateral NAc microinjections of the 5-HT4 receptor antagonist RS39604 (0.3, 1 and 3 nmol/side) plus vehicle comparison (10% DMSO) immediately prior to their MDMA (group 1) or saline (group 2) ip injection. Following drug administration, rats were returned to their home cages and provided with 100 g of standard rat chow and 800 g of water. Food intake, water intake and weight were recorded at 1 and 3 h following drug administration. This procedure was repeated until rats had received all four intracranial treatments, separated by at least 72 h.

Experiment 3: Locomotor activity of rats administered RS39604

A total of 12 rats were used for this experiment. Rats were randomly assigned to receive ip injections of either MDMA (group 1: $n=6$) or saline (group 2: $n=6$). Drug administration commenced at 08:00 hours. All rats received a 15 mm sham bilaterally into the NAc, followed by a saline injection (ip), and were placed into the locomotor chambers

for 3 h to acclimate them to experimental procedures the day prior to the first drug treatment. At this time, each group received bilateral NAc microinjections of the 5-HT4 receptor antagonist RS39604 (0.3, 1 and 3 nmol/side) plus vehicle comparison (10% DMSO) immediately prior to their MDMA (group 1) or saline (group 2) ip injection. Following drug administration, rats were placed for 3 h in locomotor chambers. Food and water was not available during this time. This procedure was repeated until rats had received all four intracranial treatments, separated by at least 72 h.

Surgeries

Under isoflourane gas anesthesia, rats undergoing microinjection procedures were stereotaxically implanted with bilateral intracranial stainless steel cannulae (26-gauge, 14 mm) 1 mm above the NAc using coordinates from Paxinos and Watson (1998): anterior/posterior=1.3, medial/lateral=1.5 and dorsal/ventral=-6.5 from bregma with the nosebar set at -3.3 mm. Cannulae were secured in place using cranioplastic cement (Vertex, Dentimex, Zeist, Holland), which was bonded to four stainless steel screws (Small Parts, USA) drilled into the skull. Stainless steel wire obturators (0.2 mm in diameter and bent at 14 mm) were inserted into the cannulae to maintain patency. During surgery and for two subsequent days, rats were treated with the analgesic Flunixin (2.5 mg/kg s.c; Troy Laboratories, Victoria, Australia). After surgery, rats recovered in a warm recovery chamber and were then returned to a clean home cage. They were allowed to recover for 7–10 days before the commencement of microinjection procedures.

Intracranial microinjections

At least 7 days following surgery, sham intracranial microinjections were performed bilaterally into the NAc using two 33-gauge stainless steel injectors bent to 15 mm in length (1 mm longer than cannulae) in order to enter the NAc. At least 24 h later, rats underwent microinjection procedures for their experiment. Each injector was attached to a 1- μ l Hamilton syringe driven by a microinjection pump (KDScientific, MA, USA). A microinjection of 0.5 μ l was delivered bilaterally to the NAc over a period of 1 min (total 1 μ l per brain). Injectors were then kept in place for a further 30 s to allow complete infusion of the drug solution from the injectors. Each rat received four counterbalanced microinjections, with a minimum of 72 h in between injections. The 5-HT4 receptor antagonist, RS39604 (Tocris Bioscience, UK), was used for the microinjection treatment. A dose-response curve was explored such that three doses of RS39604 (0.3, 1 and 3 nmol/side), plus vehicle comparison (10% DMSO), were

administered to each rat in sequences varied according to a latin square design.

Locomotor apparatus

Eight standard operant chambers [250 (depth)×310 (width)×500 (height) mm] were used. The chambers had aluminum sides and tops, while the front and back walls were made of Plexiglas. The floor was constructed of 16 metal rods (diameter 6 mm) spaced 15 mm apart. The operant chambers were housed in sound-attenuating boxes [600 (depth)×580 (width)×670 (height) mm] equipped with a fan to provide ventilation and mask noise. To detect locomotor activity, each chamber had two passive infrared (PIR) detectors (Quantum PIR motion sensor, part no. 890-087-2, NESS Security Products, Australia) positioned in the centre of each side wall 30 mm above the floor. The PIR detectors were capable of detecting small movements of the rats' head and body. The counts from the detectors were recorded by a Macintosh computer running WorkbenchMac software for data acquisition (McGregor et al. 1996).

Drugs

MDMA was purchased from the Australian Government Analytical Laboratories (AGAL) and was dissolved in 0.9% saline (CH, Australia). The 5-HT₄ receptor antagonist RS39604 was purchased from Tocris (MO, USA) and diluted in 10% dimethylsulfoxide (DMSO, Sigma USA) in 0.9% saline to the highest dose of antagonist.

Histology

At the completion of each intracranial microinjection experiment, rats were deeply anaesthetised using ip injections of pentobarbitone sodium (100 mg/kg) and transcardially perfused with saline, followed by 10% formalin for fixation of brain tissue. Brains were then removed and stored in 10% formalin for 1 week prior to sectioning. Coronal sections (60 µm thick) were then obtained from the microinjection site using a cryostat and mounted on gelatin-coated slides. Brain sections were then compared to the brain atlas of Paxinos and Watson (1998) in order to verify microinjection and cannulae placement into the NAc.

Statistical analysis

Statistical calculations were performed using SPSS version 16.1. One-way repeated measures analysis of variance (ANOVA) and independent *t* tests were used to examine differences in dependent variable measures between control

and treatment groups. The criterion for significance was set at $p < 0.05$. Where significant main effects were obtained, a priori comparisons were performed using bonferonni adjustments, with significance set at $p < 0.017$.

Results

Effect of MDMA on appetite

Figure 1 illustrates the effect of MDMA treatment (saline, 2, 5 or 10 mg/kg) on food intake, 1 and 3 h post injection; data presented are not cumulative. At 1 h following drug administration, a significant difference was found between saline and 5 mg/kg MDMA, $t(9)=2.927$, $p=0.017$, and 10 mg/kg MDMA, $t(9)=3.315$, $p=0.009$. At 3 h following drug administration, a significant difference was found between saline compared to 2 mg/kg MDMA, $t(9)=-3.806$, $p=0.004$, and 10 mg/kg MDMA, $t(9)=4.781$, $p=0.001$.

Effect of RS39604 administration on the appetite suppressant effects of MDMA

Food intake was significantly decreased at 1 h, $t(23)=9.832$, $p=0.000$, and 3 h, $t(23)=7.110$, $p=0.000$, following MDMA administration compared to saline (Fig. 2; data not cumulative). At 1 h following drug administration, food intake of MDMA-treated animals was found to be significantly higher following administration of RS39604 1 nmol compared to vehicle, $t(11)=-3.626$, $p=0.004$, and RS39604 3 nmol compared to vehicle, $t(11)=3.026$, $p=0.012$. Similarly, at 3 h following drug administration, food intake of MDMA-treated animals was found to be significantly higher following administration of RS39604 1 nmol compared to vehicle, $t(11)=-3.924$, $p=0.002$, and RS39604 3 nmol

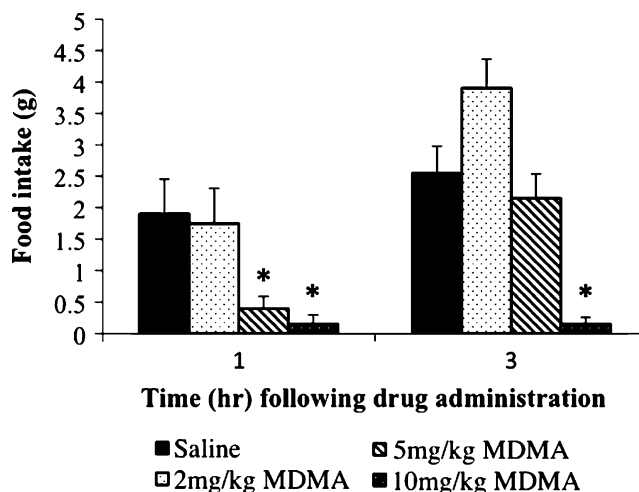


Fig. 1 Dose response curve for the effect of MDMA (ip) administration on food intake measured 1 and 3 h post injection ($*p < 0.017$)

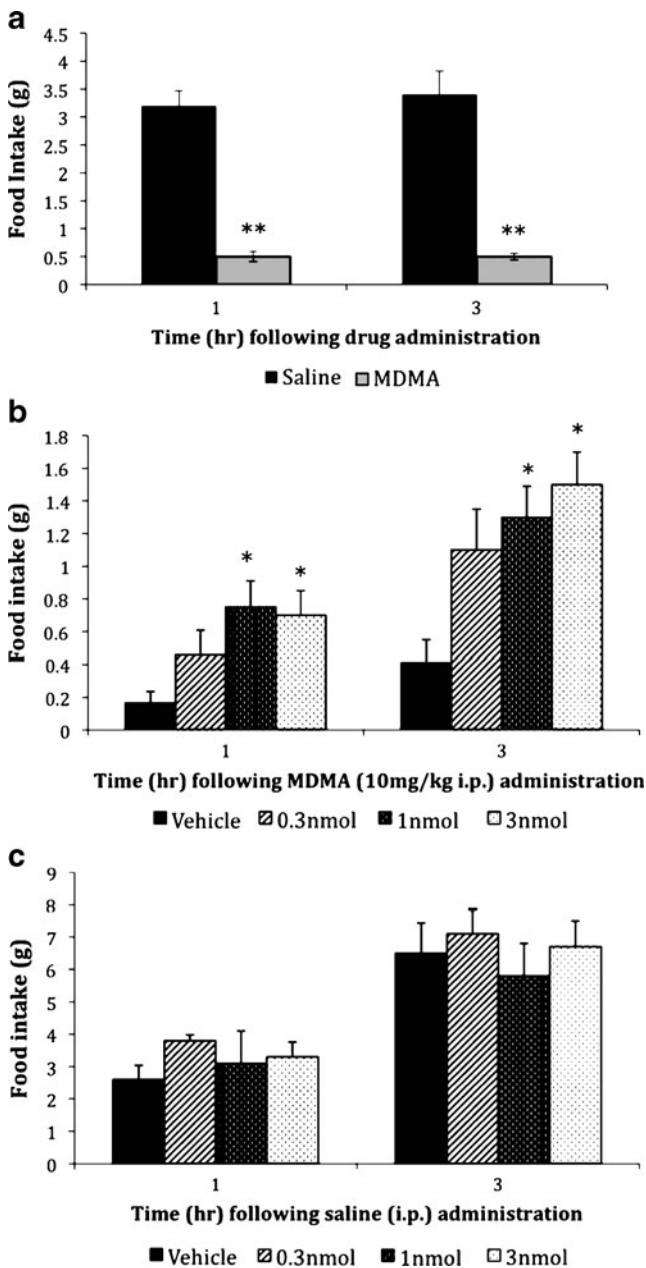


Fig. 2 Effect of MDMA (10 mg/kg ip) and RS39604 (0.3, 1, 3 nmol ic) on food intake. **a** MDMA significantly reduces food intake compared to saline at 1 and 3 h post injection (** $p < 0.01$), **b** RS39604 administration (1 nmol and 3 nmol) significantly attenuates food intake in MDMA treated rats (* $p < 0.017$), **c** no significant effect of RS39604 treatment on food intake was found in saline treated rats

compared to vehicle, $t(11) = -4.056$, $p = 0.002$. Additionally, increased food intake of MDMA-treated animals following administration of 0.3 nmol RS39604 was shown to approach significance, $t(11) = -2.680$, $p = 0.021$. No significant effect of RS39604 administration was found in saline-treated animals; however, 1 h following drug administration, food intake of saline-treated animals following administration of 0.3 nmol RS39604 was shown to approach significance, $t(11) = -2.539$, $p = 0.028$.

Effect of RS39604 administration on water intake of MDMA-treated animals

Water intake was significantly decreased at 1 h, $t(22) = 6.704$, $p = 0.000$, and 3 h, $t(22) = 5.437$, $p = 0.000$, following MDMA administration compared to saline (Fig. 3; data not cumulative). No significant effect of RS39604 treatment on water intake was obtained for the saline or MDMA-treated groups at 1 or 3 h post drug administration.

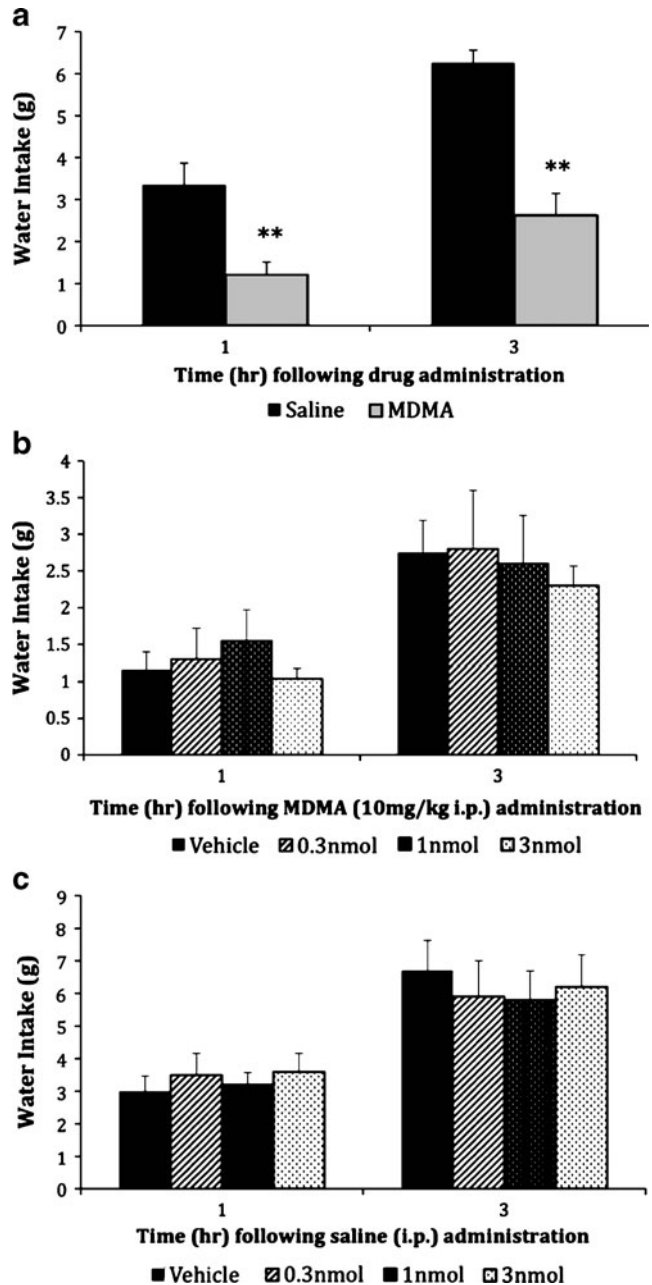


Fig. 3 Effect of MDMA (10 mg/kg ip) and RS39604 (0.3, 1, 3 nmol ic) on water intake. **a** MDMA significantly reduces water intake compared to saline at 1 and 3 h post injection (** $p < 0.01$), **b–c** no significant effect of RS39604 treatment on water intake was found in MDMA- or saline-treated rats

Effect of RS39604 administration on body weight of MDMA-treated animals

Body weight was significantly decreased (from baseline measurements recorded immediately prior to drug administration) at 1 h, $t(22)=11.9, p=0.000$, and 3 h, $t(22)=8.6, p=$

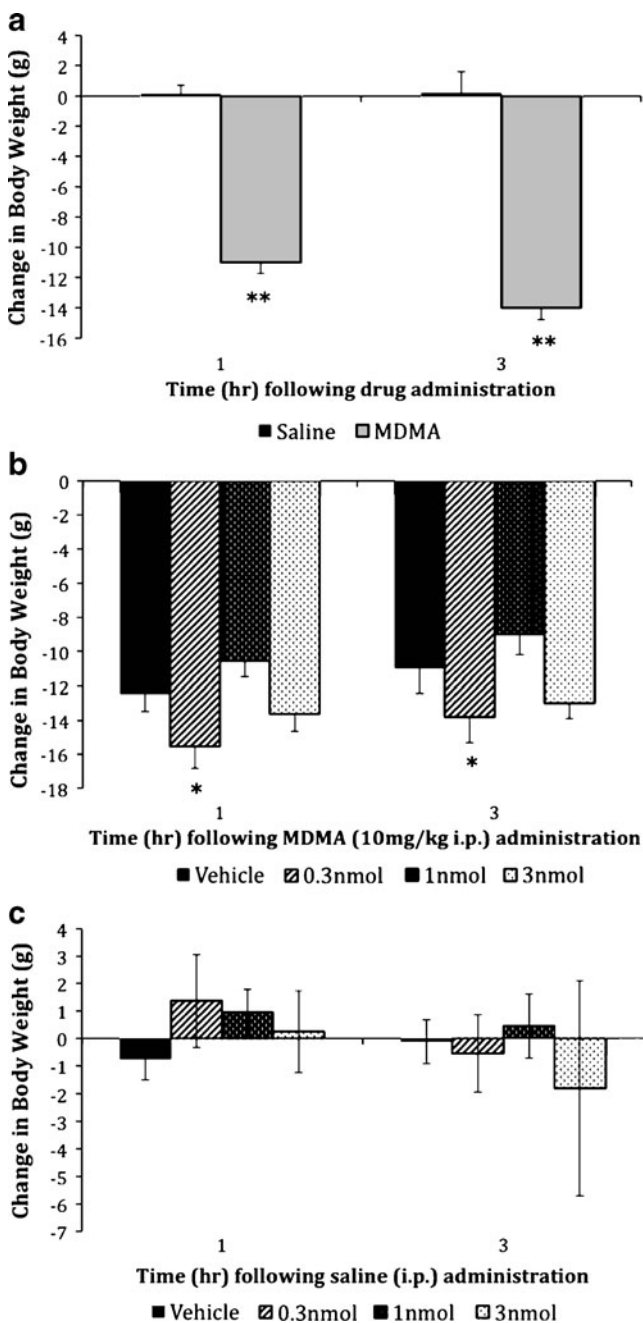


Fig. 4 Effect of MDMA (10 mg/kg ip) and RS39604 (0.3, 1, 3 nmol ic) on body weight. **a** MDMA significantly reduces body weight compared to saline at 1 and 3 h post injection (** $p<0.01$). **b** RS39604 administration (0.3 nmol) significantly increases weight loss in MDMA treated rats (* $p<0.017$). **c** no significant effect of RS39604 treatment on body weight was found in saline-treated rats

0.000, following MDMA administration compared to saline (see Fig. 4). Body weight of MDMA-treated animals was found to be significantly lower following administration of RS39604 0.3 nmol compared to vehicle at 1 h following drug administration, $t(11)=6.477, p=0.000$, and 3 h following drug administration, $t(11)=7.720, p=0.000$. No significant effect of RS39604 administration was found in saline-treated animals. See Fig. 4 for an illustration of these results.

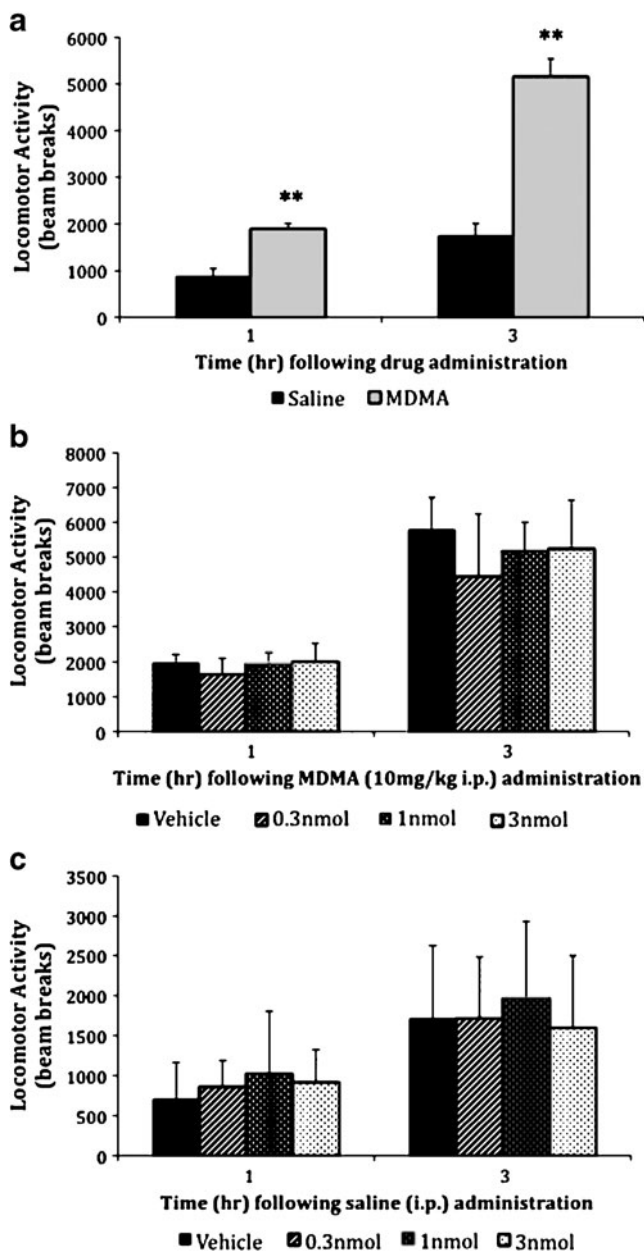


Fig. 5 Effect of MDMA (10 mg/kg ip) and RS39604 (0.3, 1, 3 nmol ic) on locomotor activity. **a** MDMA significantly increased locomotor activity compared to saline at 1 and 3 h post injection (** $p<0.01$). **b** –**c** no significant effect of RS39604 treatment on locomotor activity was found in MDMA- or saline-treated rats

Effect of RS39604 administration on locomotor activity of MDMA-treated animals

Locomotor activity was significantly decreased at 1 h, $t(10)=-4.9$, $p=0.001$, and 3 h, $t(10)=-7.2$, $p=0.000$, following MDMA administration compared to saline (see Fig. 5; data not cumulative). No significant effect of RS39604 treatment on locomotor activity was obtained for the saline- or MDMA-treated groups at 1 or 3 h following drug administration. Microinjection sites into the nucleus accumbens for Experiment 2 and 3 are shown in Fig. 6.

Discussion and conclusions

Previous research has indicated a likely role for NAc 5-HT4 receptors in the appetite-suppressant effects of MDMA in food-deprived mice (Jean et al. 2007). The findings of the present study extend this body of literature, demonstrating that direct 5-HT4 receptor antagonism in the NAc attenuates the effect of MDMA on food intake but does not attenuate the effect of MDMA on water intake, weight change or locomotor activity in rats with unrestricted access to food and water.

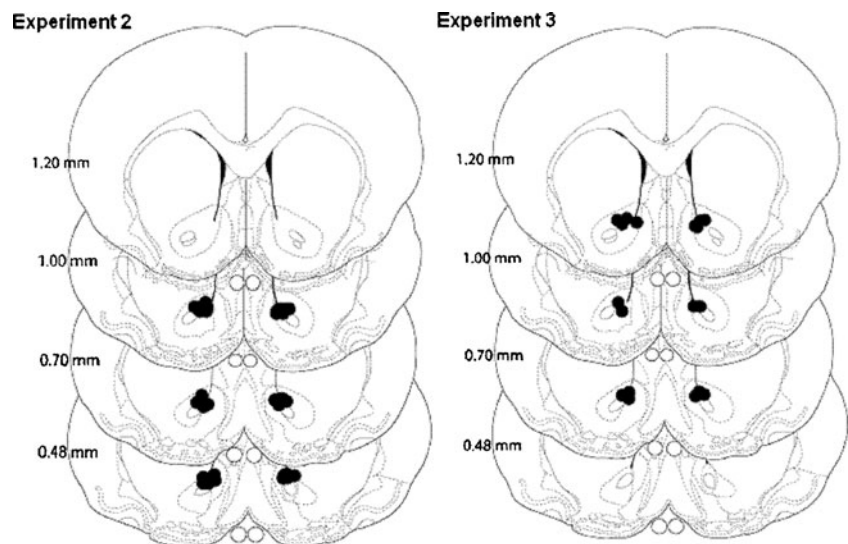
The appetite suppression observed following administration of treatments that increase serotonin release, such as fenfluramine (Lucas et al. 1998; Vickers et al. 1999) and MDMA (Frith et al. 1987; Rochester and Kirchner 1999), indicates a role for the serotonergic system in feeding behaviour. Previous research has implicated the 5-HT1 and 5-HT2 receptor subtypes in the regulation of food intake (Dourish et al. 1985; Clifton and Kennett 2006); however, the importance of other 5-HT receptor subtypes in appetite regulation should also be identified. Together with previous research demonstrating that there is an abundance of 5-HT4

receptors in the NAc (Domenech et al. 1994) and 5-HT4 receptor knockout mice display a reduction in MDMA induced appetite suppression (Jean et al. 2007), the present study shows that 5-HT4 receptors in the NAc are specific mediators of MDMA-induced appetite suppression in rats.

No effect of 5-HT4 receptor antagonism in the NAc was observed on basal feeding activity of control rats in the present study, indicating that 5-HT4 receptors do not affect appetite in general, rather that 5-HT4 receptors in the NAc are specifically involved in the appetite-suppressant effects of MDMA. This finding, however, is not consistent with previous research demonstrating an increase in basal food intake following 5-HT4 receptor antagonism in the NAc (Jean et al. 2007). Two differences in experimental proceedings may account for these discrepant findings. Firstly, as antagonist administration in the present study was performed at the commencement of the rats' dark cycle and the majority of the daily food consumption of rats occurs in the nocturnal phase (Cooper and Al-Naser 2006), a ceiling effect may have been observed in the present study. Supporting this, 5-HT4 antagonism was found by Jean et al. (2007) to elevate food intake in fed mice but not food-deprived, motivated mice. Secondly, the study by Jean et al. (2007) used mice for their rodent model of feeding behaviour, indicating a possible species difference in the role of 5-HT4 receptors in the NAc in basal feeding behaviour. The present results, however, that 5-HT4 receptor antagonism in the NAc had no effect on basal feeding activity, suggest that these receptors are not tonically activated under basal conditions and are distinctly involved in alterations in feeding behaviour elicited by MDMA administration.

To test the possibility that the 5-HT4 receptor exerted its effects on food intake through non-specific mechanisms, water intake was also measured in the present study. Our

Fig. 6 Location of microinjection sites into the NAc for experiment 2 ($n=24$) and experiment 3 ($n=12$). Sections were adapted from the brain atlas of Paxinos and Watson (1998). Numbers indicate distance (mm) from bregma



study demonstrated that MDMA administration resulted in decreased water intake, consistent with previous findings that rats trained to lever press for water display reductions in water intake following MDMA administration (Laraway et al. 2003). This effect, however, was not found to be attenuated by 5-HT₄ receptor antagonism in the NAc, indicating that MDMA inhibits feeding and drinking behaviours through separate mechanisms.

The finding that MDMA administration resulted in an increase in locomotor activity is consistent with subjective reports in human studies (Parrott 2001) and observations in animal studies (Gold et al. 1988; Spanos and Yamamoto 1989) of increased locomotor activity following MDMA administration. Of the many serotonin receptor subtypes, the 5-HT₄ receptor was hypothesised to be involved in locomotor activity due the high density of this receptor in the NAc, a key brain region mediating locomotor effects of psychostimulants (Reavill et al. 1998). The results of this study, however, do not support this hypothesis as 5-HT₄ receptor antagonism in the NAc was not found to attenuate the locomotor effects of MDMA.

The finding that 5-HT₄ receptor antagonism attenuated the effects of MDMA on food intake but not on locomotor activity suggests that MDMA specifically reduces food intake, which is not a consequence of increased motor stimulant behaviour to prevent food consumption. These results also imply that different mechanisms underlie the effect of MDMA on food intake and locomotor activity. While the present study does not provide evidence that 5-HT₄ receptors in the NAc are involved in the locomotor effects of MDMA, previous research suggests an important role for serotonin in MDMA-induced locomotor activity (Callaway et al. 1990; Callaway et al. 1991). The serotonin receptor subtypes that have been demonstrated to influence the locomotor effects of MDMA include the 5-HT_{1B}, 5-HT_{2A} and 5-HT_{2C} receptor subtypes (Fletcher et al. 2002, Fletcher et al. 2006). While 5-HT₄ receptors in the NAc do not seem to be involved in MDMA-induced locomotor activity, they do mediate cocaine-induced locomotor activity (McMahon and Cunningham 1999). The results of the present study add to a growing body of literature aiming to characterise the specific receptor subtypes involved in the locomotor effects of MDMA.

This study provides evidence for a significant decrease in weight observed in the 3 h following MDMA administration. MDMA-induced weight loss occurring in this limited time frame is most likely due to increased micturition, defaecation, ejaculation or sweating. While none of these outputs were measured in the present study, a previous conditioned place preference report measured weight loss associated with an increase in seminal plugs, urination and defaecation for 3 h following MDMA administration (Bilsky et al. 1991). The effect of low dose administration of the 5-HT₄ antagonist into

the NAc to significantly enhance MDMA-induced weight loss is a surprising finding, particularly when considering that this dose had no effect on any other measure. The mechanism for this effect is currently unknown, yet may be a presynaptic effect on other neurotransmitter (such as dopamine) release to enhance MDMA-induced weight loss (Barnes and Sharp 1999). While this study and that of Bilsky et al. (1991) both show an immediate effect of MDMA to reduce body weight, subjective measures of immediate weight changes in human users have not yet been reported. Future research may consider determining whether acute weight loss is a factor that immediately reinforces continued use and whether these effects on weight change are perceived as enduring in MDMA users.

The results of the present study show that direct antagonism of 5-HT₄ receptors in the NAc are involved in the appetite suppressant effects of MDMA, but do not mediate MDMA-induced water intake or locomotor activity, and that MDMA has acute effects on body weight. Our results confirm the role of NAc 5-HT₄ receptors in MDMA-induced regulation of food intake and demonstrate the specificity of this effect in animals with unrestricted access to food and water. Further, any effect of 5-HT₄ receptor antagonism in the NAc to reduce MDMA food intake is not related to changes in locomotor activity. Our findings contribute to understanding the neurobiology of MDMA administration and may aid in the development of successful treatment plans targeted at young adult users.

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