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Δ9-Tetrahydrocannabinol Induces Dopamine Release in the Human Striatum

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The influence of cannabis on mental health receives growing scientific and political attention. An increasing demand for treatment of cannabis dependence has refueled the discussion about the addictive potential of cannabis. A key feature of all addictive drugs is the ability to increase synaptic dopamine levels in the striatum, a mechanism involved in their rewarding and motivating effects. However, it is currently unknown if cannabis can stimulate striatal dopamine neurotransmission in humans. Here we show that Δ9-tetrahydrocannabinol (THC), the main psychoactive component in cannabis, induces dopamine release in the human striatum. Using the dopamine D_2/D_3 receptor tracer [^{11}C]raclopride and positron emission tomography in seven healthy subjects, we demonstrate that THC inhalation reduces [^{11}C]raclopride binding in the ventral striatum and the precommissural dorsal putamen but not in other striatal subregions. This is consistent with an increase in dopamine levels in these regions. These results suggest that THC shares a potentially addictive property with other drugs of abuse. Further, it implies that the endogenous cannabinoid system is involved in regulating striatal dopamine release. This allows new directions in research on the effects of THC in neuropsychiatric disorders, such as schizophrenia.
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INTRODUCTION

The debate whether cannabis can be characterized as an addictive drug has been ongoing for many years (Grinspoon *et al*, 1997). Recently, a substantial increase in the demand for treatment of cannabis dependence has intensified this discussion (Hall, 2006). The rewarding properties of addictive drugs are thought to be mediated by their action on the mesolimbic dopamine system (for review see Wise, 2004; Hyman *et al*, 2006). This dopamine system originates in the ventral tegmental area and projects to the ventral striatum, which predominantly comprises the nucleus accumbens. Addictive drugs probably induce their rewarding effects by enhancement of synaptic dopamine levels in the ventral striatum (Wise, 2004; Hyman *et al*, 2006). In the human striatum, increased dopamine levels have been found with the use of neuroimaging techniques after the administration of amphetamine (Laruelle *et al*, 1996; Breier *et al*, 1997; Drevets *et al*, 2001; Martinez *et al*, 2003, 2007), cocaine (Schlaepfer *et al*, 1997), alcohol (Boileau *et al*, 2003), and nicotine (Brody *et al*, 2004, 2006). In animals, it has been demonstrated that cannabinoid substances such as Δ9-tetrahydrocannabinol (THC), the main psychoactive component in cannabis (Gaoni and Mechoulam, 1964), also stimulate striatal dopamine neurotransmission (for review see Tanda and Goldberg, 2003; Lupica *et al*, 2004; Gardner, 2005). Cannabinoids enhance neuronal firing of mesolimbic dopamine neurons (French, 1997; French *et al*, 1997; Gessa *et al*, 1998) and elevate striatal dopamine levels (Ng Cheong Ton *et al*, 1988; Chen *et al*, 1990; Tanda *et al*, 1997; Malone and Taylor, 1999; Fadda *et al*, 2006), both through activation of cannabinoid CB1 receptors (French, 1997; French *et al*, 1997; Tanda *et al*, 1997; Gessa *et al*, 1998; Malone and Taylor, 1999). However, whether THC affects the human striatal dopamine system is currently unknown.

The purpose of the present study was to investigate whether THC can induce dopamine release in the striatum of healthy human subjects. This was assessed using positron emission tomography (PET) and the dopamine D_2/D_3 receptor ligand [^{11}C]raclopride. With this method, an increase in striatal synaptic dopamine concentrations can be determined by a reduction in [^{11}C]raclopride binding (Breier *et al*, 1997; Laruelle *et al*, 1997). On the basis of findings from animal studies, our hypothesis was that THC should reduce [^{11}C]raclopride binding in the human striatum, consistent with striatal dopamine release.

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MATERIALS AND METHODS

Subjects

Healthy male subjects were recruited through advertisements on the Internet. All subjects had a history of mild cannabis use for at least 1 year, defined as using cannabis more than four times a year and at most once a week. In addition, it was required that they never experienced psychotic effects after cannabis use and did not meet criteria for 'Paranoid Ideation' or 'Psychoticism' on the self-report symptom checklist SCL-90 (Derogatis, 1983). Mild cannabis users were selected as they *a priori* could be expected to tolerate the THC challenge used in this experiment although not having long-term effects associated with frequent cannabis use. All subjects were in good physical health as assessed by medical history, physical examination, electrocardiogram (ECG), and routine laboratory tests. Urine screening for cannabis, amphetamine, cocaine, and morphine was performed at screening and on both study days. Subjects with a positive drug test on other drugs than cannabis were excluded from the study. Subjects with a positive cannabis test at screening were tested again, and were required to be negative before the first study day. Subjects were excluded from participation in case of history of alcohol or drug abuse and in case of major current psychiatric diagnosis. In addition, subjects were excluded if they, or a first- or second-degree relative, had a lifetime history of a clinically significant psychiatric or neurological illness. Use of medication at the time of the study was not allowed. All volunteers gave written informed consent before entry into the study. The study was approved by the Medical Research Ethics Committee of the University Medical Center Utrecht, the Netherlands.

Design and Procedure

In a double-blind, randomized, placebo-controlled, cross-over study, subjects had two PET scans after either administration of THC or placebo. Scanning sessions were separated by at least 2 weeks to allow for complete clearance of drug between both occasions. Subjects arrived 2 h before the start of the scanning procedure at the Department of Nuclear Medicine and PET Research of the VU University Medical Center in Amsterdam, the Netherlands, having fasted for at least 4 h before their arrival. Subjects refrained from cannabis for at least 2 weeks before the first study day until study completion and from alcohol for 24 h before each study day. Caffeine intake and smoking were not allowed on study days. Use of drugs of abuse, including cannabis, was checked with urine drug screenings, which had to be negative on the day of the PET scans. Use of alcohol, caffeine, and nicotine was checked by self-report. A standard breakfast or lunch was served and venous catheters were placed in each arm, one for [^{11}C]raclopride infusion and the other for venous blood sampling.

Drugs and Administration

Preparation and administration of drugs was performed according to Zuurman *et al* (2008). THC was purified from *Cannabis sativa* according to GMP-compliant procedures (Farmalyse BV, Zaandam, the Netherlands) and was dissolved in 200 μl 100 vol% alcohol. The solvent was used as placebo. Drugs were administered using a Volcano® vaporizer (Storz–Bickel GmbH, Tuttlingen, Germany). Five minutes before administration, 8 mg of THC was vaporized into an opaque polythene bag equipped with a valved mouthpiece, preventing the loss of THC in between inhalations. Subjects inhaled the volume of this bag in three or four subsequent breaths, holding their breath for 10 s after each inhalation. They were not allowed to speak during the inhalation process, which was practiced at screening using placebo.

Production of [^{11}C]raclopride

[^{11}C]Raclopride was synthesized by methylation of *O*-desmethyl raclopride (obtained from ABX, Radeberg, Germany) with $^{11}CH_3I$ in dimethylsulfoxide at 80°C for 5 min, utilizing a Nuclear Interface methylation synthesis module. The resulting product was purified from the reaction mixture by HPLC (μbondapak 7.8 × 300; 0.01 M H_3PO_4/MeCN 70 : 30, 5 ml/min, UV at 254 nm). The collected fraction containing [^{11}C]raclopride was diluted with 40 ml of 1 mM NaOH in water and this mixture was subsequently passed over a tC18 SepPak. After washing the SepPak with 20 ml of water for injection, [^{11}C]raclopride was eluted from the SepPak with 1.2 ml of sterile ethanol and a sterile solution of NaH_2PO_4 in saline (7.1 mM, pH 5.4). The final solution was transferred to a sterile product vial by a sterile 0.22 μm MIllex GV filter, yielding a sterile, pyrogen free solution of [^{11}C]raclopride with a (radio)-chemical purity of >98% whereas the specific activity ranged from 26 to 104 GBq/μmol at time of injection. The complete production procedure was performed in accordance with the EU guideline 'Eudralex volume 4: Good Manufacturing Practices' and were approved by the Dutch health authorities (license number 107627A).

Positron Emission Tomography

PET scans were performed on an ECAT EXACT HR+ scanner (Siemens/CTI, Knoxville, TN, USA), which is located at the Department of Nuclear Medicine and PET Research of the VU University Medical Center in Amsterdam, the Netherlands. This scanner enables the acquisition of 63 transaxial planes over a 15.5 cm axial field of view, thus allowing the whole brain to be imaged.

Five minutes after inhalation of placebo or THC, [^{11}C]raclopride was given as a bolus plus constant infusion. [^{11}C]Raclopride was delivered in a 50 ml volume and administered by a computer operated infusion pump (Med-Rad, Beek, the Netherlands). First, a bolus dose of 28 ml was given over 3.1 min, followed by constant infusion of 22 ml at 0.15 ml/h for 88 min. Thus, the bolus to infusion ratio (K_{bol}) was 112 min (Carson *et al*, 1997). A 40 min scanning period with eight successive frames of 5 min was initiated 40 min after the start of [^{11}C]raclopride administration. Finally, a transmission scan of 10 min was performed to correct for photon attenuation. Correction for emission contamination was performed using the dwell profile method (van der Weerdt *et al*, 2004).

Image Reconstruction

All PET sinograms were corrected for dead time, decay, randoms, scatter, and tissue attenuation. All PET emission scans were reconstructed with FORE + 2D FBP using a 0.5 Hanning filter, resulting in a transaxial spatial resolution of ~7 mm in the center of the field of view. Images were then transferred to workstations (Sun Microsystems, Santa Clara, CA, USA) for further analysis.

Behavioral, Subjective, and Physiological Measurements

Behavioral ratings were assessed with the 18-item Brief Psychiatric Rating Scale (BPRS; Overall and Gorham, 1962). This structured interview was performed at baseline and 21 and 102 min after THC administration. Total BPRS scores were analyzed together with scores for the factors thinking disorder (BPRS items: conceptual disorganization, hallucinatory behavior, and unusual thought content), withdrawal–retardation (BPRS items: blunted affect and emotional withdrawal), anxiety–depression (BPRS items: anxiety, guilt feelings, and depressive mood), and hostility–suspiciousness (BPRS items: hostility, suspiciousness, and uncooperativeness) (Hedlund and Vieweg, 1980).

A rating scale consisting of 16 visual analogue scales was used to determine subjective effects. From these analogue scales three factors were calculated, corresponding to alertness, contentedness, and calmness (Bond and Lader, 1974). Psychedelic effects were assessed using an adapted version of a 13-item visual analogue rating scale, originally described by Bowdle and colleagues (Bowdle *et al*, 1998; Zuurman *et al*, 2008). The visual analogue scale (VAS) 'feeling high' was analyzed individually and composite scores of 'external perception' and 'internal perception' were calculated (Zuurman *et al*, 2008). Both rating scales were performed consecutively at baseline and 7, 12, 17, 32, and 100 min after THC administration.

ECG was monitored continuously whereas blood pressure and heart rate were measured at baseline and 6, 11, 15, 19, 34, 49, 64, 79, and 94 min after start of THC administration.

Blood Sampling

Venous blood samples were collected to determine plasma concentrations of THC and its two most important metabolites, 11-OH-THC and 11-nor-9-carboxy-THC. Blood samples were withdrawn 5, 10, 20, 35, 55, and 90 min after THC administration and processed according to Zuurman *et al* (2008). Additional venous blood samples were withdrawn 40, 60, 70, and 80 min after start of [^{11}C]raclopride administration to measure [^{11}C]raclopride metabolism. These samples were processed according to Schuit *et al* (2007).

Magnetic Resonance Imaging

Magnetic resonance imaging (MRI) of all subjects was performed at the Department of Radiology of the University Medical Center in Utrecht, the Netherlands, for anatomical definition. MRI scans were acquired using a 1.5 T scanner (Philips Gyroscan; Philips Medical Systems, Best, the Netherlands). T1-weighted, 3D, fast field echo scans with 160–180 1.2 mm contiguous coronal slices (echo time, 4.6 ms; repetition time, 30 ms; flip angle 301; field of view 256 mm; Hulshoff Pol *et al*, 2001) were used.

Regions of Interest

All MRI scans were rotated to acquire horizontal lines between anterior and posterior commissures (AC–PC line) in the sagittal plane. Then the striatum was divided into five anatomical regions of interest (ROI; see Figure 1; Table 1), according to published criteria (Mawlawi *et al*, 2001; Martinez *et al*, 2003). These ROIs were delineated on MRI scans oriented in the coronal plane using DISPLAY. ROIs were defined for ventral striatum, precommissural dorsal caudate, precommissural dorsal putamen, postcommisural caudate, and postcommissural putamen. These ROIs were classified into three functional subdivisions: limbic striatum (ventral striatum), associative striatum (consisting of precommissural dorsal caudate, precommissural dorsal putamen, and postcommisural caudate), and sensorimotor striatum (postcommissural putamen; Mawlawi *et al*, 2001; Martinez *et al*, 2003). Cerebellar hemispheres were also defined on the MRI scans. After reconstruction, individual PET frames were co-registered to the first frame to correct for motion and summed over all frames. These summed PET images were co-registered to the rotated MRI scan using VINCI software (Cizek *et al*, 2004). After projection of the ROIs on the co-registered summed PET images, activity was calculated for each ROI as the volume weighted average of left and right regions. Activity in associative striatum was derived as the volume weighted average of precommissural dorsal caudate, precommissural dorsal putamen, and postcommisural caudate, whereas activity in striatum as a whole was calculated as the volume weighted average of all five ROIs (Table 1).

Outcome Measures

Nondisplaceable binding potential (BP_{ND}; Innis *et al*, 2007) was used as measure of dopamine D_2/D_3 receptor availability. BP_{ND} was defined as the distribution volume ratio (DVR) minus 1 (Lammertsma *et al*, 1996). As scans were performed during steady state, DVR could be obtained using the average activity concentration in a ROI divided by that of the cerebellum ROI, which was used as reference. In this way, BP_{ND} was calculated for all ROIs for both scanning sessions.

Statistical Analysis

Group differences in BP_{ND} between placebo and THC were analyzed using repeated measures ANOVA with ROI and drug as factors. *Post hoc* analysis was performed for each ROI using paired *t*-tests. Behavioral, subjective, psychedelic, and physiological effects were corrected for baseline values and also analyzed using repeated measures ANOVA with drug and time as factors. *Post hoc* analysis was performed using paired *t*-tests. Therefore, a mean score was calculated for each parameter and compared between placebo and THC. Differences in PET scan parameters and [^{11}C]raclopride concentrations were measured using paired *t*-tests.

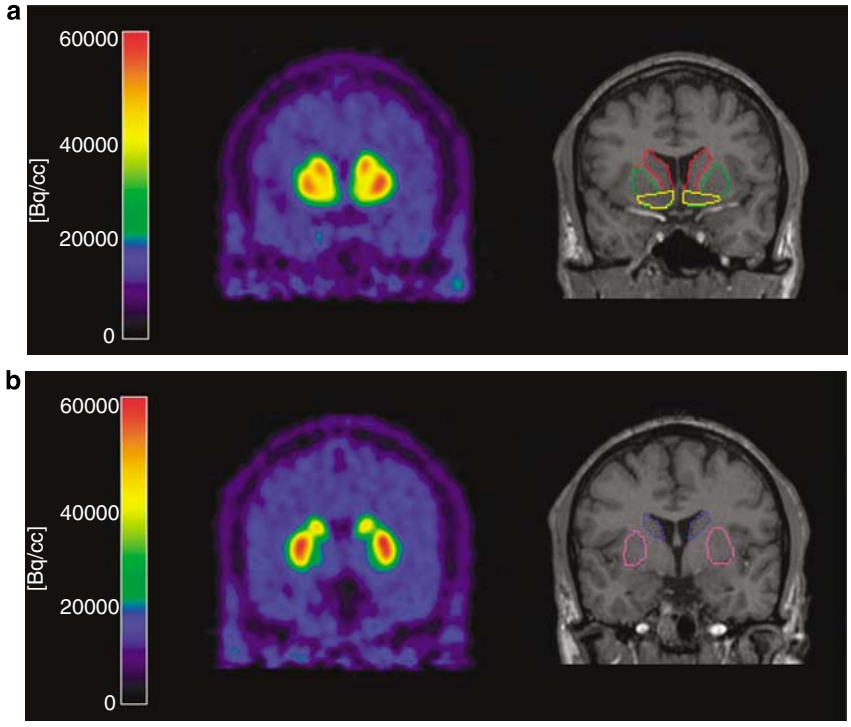

Figure 1 Coronal slices of (left) PET [^{11}C]raclopride and (right) co-registered magnetic resonance imaging (MRI) scans at the level of the striatum, (a) anterior and (b) posterior to the *AC*-plane. Striatal subregions are indicated on the MRI scans: ventral striatum (yellow), precommissural dorsal putamen (green), precommissural dorsal caudate (red), postcommissural putamen (purple), and postcommissural caudate (blue).

Table 1 Effects of Δ9-Tetrahydrocannabinol (THC; 8 mg) on [^{11}C]raclopride Nondisplaceable Binding Potential (BP_{ND}), Reflecting Dopamine D_2/D_3 Receptor Availability (mean ± SD; $n = 7$)

Region	BP_{ND} placebo	BP_{ND} THC	Difference (%)	*p*-values
Ventral striatum	1.40 ± 0.24	1.35 ± 0.24	−3.43 ± 3.70	0.029*
Precommissural dorsal caudate	2.18 ± 0.25	2.12 ± 0.13	−2.09 ± 6.44	0.355
Precommissural dorsal putamen	2.75 ± 0.24	2.64 ± 0.16	−3.88 ± 4.07	0.042*
Postcommissural caudate	1.62 ± 0.19	1.55 ± 0.15	−4.12 ± 7.14	0.157
Postcommissural putamen	2.74 ± 0.29	2.69 ± 0.20	−1.50 ± 4.42	0.329
Striatum	2.28 ± 0.22	2.21 ± 0.12	−2.57 ± 4.42	0.153

Striatum values were calculated as the volume weighted averages of all five regions of interest. *BP significantly different between THC and placebo.

A *p*-value less than 0.05 was considered statistically significant.

RESULTS

Subjects

Nine healthy male subjects gave informed consent for this study. Seven volunteers completed the study procedure. One subject was excluded because of positive urine drug screening on the first study day. Another subject did not complete the second scanning session because of anxiety. Mean age of the seven subjects was 21.9 ± 2.7 years (range 20–27). Mean height, weight, and BMI were 183 ± 8 cm (range 172–196), 80 ± 9 kg (range 72–98), and 23.8 ± 0.9 kg/m^2 (range 22.9–25.5), respectively. All subjects were familiar with the effects of cannabis. Two subjects used cannabis less than once a month, one subject three times a month, three subjects twice a month and one subject used cannabis once a week. They all showed negative urine screening at both study days.

PET Scan Parameters

Mean injected dose of [^{11}C]raclopride was similar between placebo (770 ± 30 MBq) and THC sessions (810 ± 90 MBq; $p = 0.254$). In addition, total mass of administered raclopride (6.1 ± 2.3 and 4.6 ± 1.5 μmol for placebo and THC sessions, respectively; $p = 0.181$) was not significantly different between sessions.

There were no significant changes in equilibrium levels of striatal activity, expressed by percentage change of activity concentration over time, between placebo (−0.10 ± 0.16% per min) and THC (−0.12 ± 0.08% per min) sessions

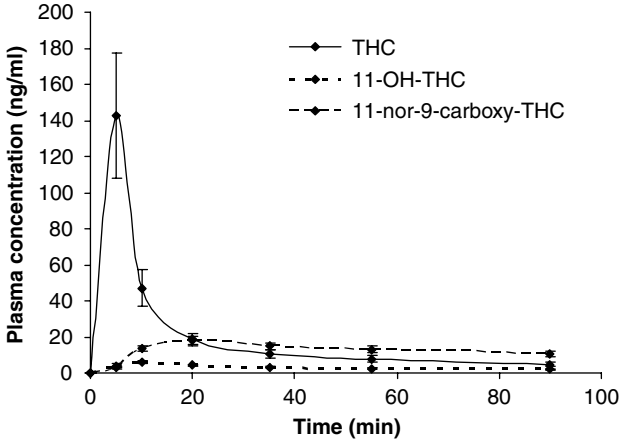

Figure 2 Plasma concentrations of Δ9-tetrahydrocannabinol (THC) and its main metabolites 11-OH-THC and 11-nor-9-carboxy-THC after inhalation of 8 mg THC (mean ± SEM; $n = 7$).

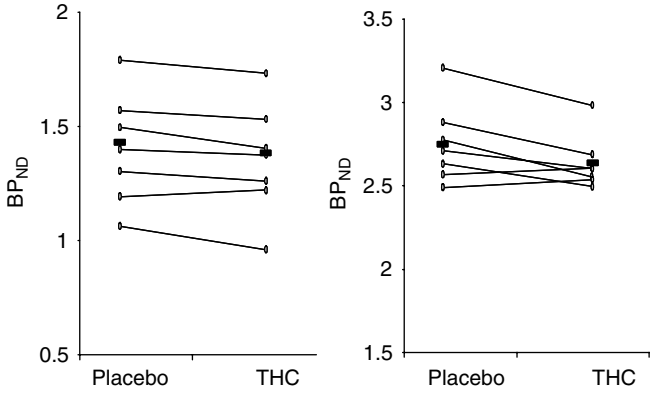

Figure 3 Effects of Δ9-tetrahydrocannabinol inhalation (8 mg) on [^{11}C]raclopride binding in (left) ventral striatum and (right) precommissural dorsal putamen of healthy subjects ($n = 7$). Data are presented as [^{11}C]raclopride nondisplaceable binding potential, reflecting dopamine D_2/D_3 receptor availability.

($p = 0.633$). In addition, the slope (ie change over time during scanning) of the ratio between striatum and cerebellum [^{11}C]raclopride concentrations was not significantly different between sessions (-0.0011 ± 0.0030 and -0.0010 ± 0.0025 for placebo and THC sessions, respectively; $p = 0.967$).

Blood Sample Analysis

THC plasma concentration reached a maximum of 143 ± 91 ng/ml 5 min after inhalation, decreasing rapidly thereafter. Plasma concentrations of the main metabolites of THC, 11-OH-THC, and 11-nor-9-carboxy-THC, peaked at 10 min (6 ± 2 ng/ml) and 20 min (18 ± 5 ng/ml) after inhalation, respectively (Figure 2).

[^{11}C]Raclopride concentrations were not significantly different between placebo and THC sessions in both whole blood (0.59 ± 0.25 and 0.61 ± 0.28 kBq/g, respectively; $p = 0.717$) and plasma (1.02 ± 0.42 and 1.06 ± 0.43 kBq/g, respectively; $p = 0.687$), normalized to the effectively injected dose. In addition, the fraction of parent [^{11}C] raclopride was not significantly different ($p = 0.191$) between placebo ($80.4 \pm 5.6\%$) and THC ($77.5 \pm 4.3\%$).

Dopamine D_2/D_3 Receptor Availability

BP_{ND} of [^{11}C]raclopride, reflecting dopamine D_2/D_3 receptor availability, was significantly reduced in the ventral striatum and the precommissural dorsal putamen after inhalation of THC compared to placebo ($-3.43 \pm 3.70\%$, $p = 0.029$ and $-3.88 \pm 4.07\%$, $p = 0.042$, respectively; Figure 3). In other subdivisions of the striatum no significant differences were found between THC and placebo (Table 1).

Behavioral, Subjective, and Physiological Measurements

Analysis of variance revealed a significant drug × time effect on the BPRS total score ($F(2,12) = 11.28$, $p = 0.002$) and on the BPRS composite score withdrawal–retardation ($F(2,12) = 18.23$, $p < 0.001$). THC-induced significant increases in VAS scores of 'feeling high' ($F(5,30) = 3.88$, $p = 0.008$), 'external perception' ($F(5,30) = 2.91$, $p = 0.029$) and 'calmness' ($F(5,30) = 3.22$, $p = 0.019$), and a decrease on 'alertness' ($F(5,30) = 3.07$, $p = 0.024$). Heart rate increased significantly after THC compared with placebo ($F(9,54) = 9.36$, $p < 0.001$). Behavioral, subjective, and physiological measurements are summarized in Table 2 and Table 3. No significant associations between measures of dopamine release and behavioral, subjective or physiological effects were demonstrated.

Table 2 Behavioral, Subjective, and Physiological Effects of Δ9-Tetrahydrocannabinol ($n = 7$)

Assessment	Drug × time interaction
BPRS total score	$F(2,12) = 11.28$, $p = 0.002$*
Thinking disorder	–
Withdrawal–retardation	$F(2,12) = 18.23$, $p < 0.001$*
Anxiety–depression	$F(2,12) = 0.11$, $p = 0.898$
Hostility–suspiciousness	$F(2,12) = 1.64$, $p = 0.235$
VAS alertness	$F(5,30) = 3.07$, $p = 0.024$*
VAS contentedness	$F(5,30) = 1.73$, $p = 0.157$
VAS calmness	$F(5,30) = 3.22$, $p = 0.019$*
VAS feeling high	$F(5,30) = 3.88$, $p = 0.008$*
VAS internal perception	$F(5,30) = 2.51$, $p = 0.052$
VAS external perception	$F(5,30) = 2.91$, $p = 0.029$*
Heart rate	$F(9,54) = 9.36$, $p < 0.001$*
Systolic blood pressure	$F(9,54) = 1.38$, $p = 0.240$
Diastolic blood pressure	$F(9,54) = 0.72$, $p = 0.689$

Statistical analysis was performed on baseline corrected data using repeated measures ANOVA with drug and time as factors. See for *post hoc* analysis Table 3.
*Significant difference between THC and placebo. BPRS, Brief Psychiatric Rating Scale; VAS, Visual Analogue Scale.

DISCUSSION

This study examined the effects of THC inhalation on [^{11}C]raclopride specific binding in seven healthy volunteers,

Table 3 *Post hoc* Analysis Performed with Paired *t*-tests of the Baseline Corrected Behavioral, Subjective, and Physiological Parameters that Demonstrated a Significant Drug × Time Effect (See Table 2) (mean ± SD; $n = 7$)

Assessment	Mean placebo score	Mean THC score	*p*-values
BPRS total score	−0.10 ± 0.16	1.95 ± 1.42	0.008*
BPRS withdrawal–retardation	0.00 ± 0.00	0.62 ± 0.23	<0.001*
VAS alertness	0.62 ± 2.23	−6.34 ± 6.31	0.038*
VAS calmness	2.11 ± 3.54	4.88 ± 8.63	0.458
VAS feeling high	0.45 ± 1.50	27.33 ± 17.73	0.009*
VAS external perception	0.17 ± 0.30	9.77 ± 10.16	0.048*
Heart rate	−4.71 ± 4.38	16.17 ± 19.87	0.040*

For each parameter a mean score was calculated and compared between THC and placebo.
*Significant difference between THC and placebo. BPRS, Brief Psychiatric Rating Scale; VAS, Visual Analogue Scale.

finding a reduction in the ventral striatum and precommissural dorsal putamen. The reduction in [^{11}C]raclopride specific binding is consistent with an increase in dopamine levels in these regions.

This is the first study demonstrating THC-induced dopamine release in the human striatum. This result is in line with animal findings, showing enhanced neuronal firing of mesolimbic dopamine neurons after administration of cannabinoids (French, 1997; French *et al*, 1997; Gessa *et al*, 1998). In addition, it is consistent with microdialysis studies demonstrating that cannabinoids induce elevated striatal dopamine levels (Ng Cheong Ton *et al*, 1988; Chen *et al*, 1990; Tanda *et al*, 1997; Malone and Taylor, 1999; Fadda *et al*, 2006). These effects are dependent on the activation of cannabinoid CB1 receptors (French, 1997; French *et al*, 1997; Tanda *et al*, 1997; Gessa *et al*, 1998; Malone and Taylor, 1999).

The ability of THC to induce dopamine release in the human striatum suggests that THC shares addictive properties with other drugs of abuse. Dopamine release in the striatum is a key feature of all addictive drugs, specifically involved in their rewarding effects and in the formation of reward related associations (Wise, 2004; Hyman *et al*, 2006). However, whereas amphetamine (Drevets *et al*, 2001; Martinez *et al*, 2003, 2007), cocaine (Schlaepfer *et al*, 1997), alcohol (Boileau *et al*, 2003), and nicotine (Brody *et al*, 2004, 2006) cause reductions in dopamine D_2/D_3 receptor availability in the range of 10–30%, we found a relatively modest THC-induced decrease of 3.4 and 3.9% in the ventral striatum and the precommissural dorsal putamen, respectively. Interestingly, this modest decrease in [^{11}C]raclopride binding is consistent with the moderate increase in striatal dopamine levels measured after administration of THC in animals (Ng Cheong Ton *et al*, 1988; Chen *et al*, 1990; Tanda *et al*, 1997; Malone and Taylor, 1999). Assuming a ratio between increase in dopamine levels and reduction in [^{11}C]raclopride binding of approximately 40 : 1 (Breier *et al*, 1997; Laruelle *et al*, 1997), our data indicate an increase in dopamine concentrations in the ventral striatum of 136%. This is in line with the THC-induced increase in striatal dopamine levels as demonstrated in microdialysis studies (Ng Cheong Ton *et al*, 1988; Chen *et al*, 1990; Tanda *et al*, 1997; Malone and Taylor, 1999).

As THC was dissolved in 100 vol% alcohol and the solvent was used as placebo, we cannot exclude that the inhalation of alcohol has caused dopamine release. However, this is very unlikely, as only 200 μl alcohol was administered. Please note that the placebo scan was subtracted from the THC scan and both conditions contained the same amount of alcohol.

THC-induced well-known behavioral, subjective, and physiological effects (D'Souza *et al*, 2004; Ilan *et al*, 2005) in our subjects, replicating effects caused by the highest dose of THC administered in previous research using the same vaporizing device (Strougo *et al*, 2008; Zuurman *et al*, 2008). In addition, THC plasma concentrations in this study were comparable with or even higher than those obtained after smoking of high-potency cannabis (Huestis *et al*, 1992; Ramaekers *et al*, 2006). Thus, our results indicate that a relatively high dose of THC induces a moderate degree of dopamine release in the human striatum. This effect may be explained by the indirect effects of THC on striatal dopamine levels through cannabinoid CB1 receptors on glutamate and GABA neurons in the nucleus accumbens and the ventral tegmental area (Schlicker and Kathmann, 2001; Lupica *et al*, 2004). Other drugs of abuse have more direct effects on the dopamine system (Koob *et al*, 1998; Hyman *et al*, 2006).

As the PET scan was performed around 45–85 min after inhalation of THC it could be argued that most of the effect of THC on dopamine release had dissipated at the time of the scan. By this time, plasma concentrations of THC were only 2.0–4.4% of the maximum concentration. However, pharmacokinetic/pharmacodynamic (PK/PD) models that have been described recently indicate that central nervous system effects of THC last much longer than suggested by the rapid decline of plasma concentrations (Strougo *et al*, 2008). Application of these PK/PD-models to this study showed that 84.5–95.9% of the maximum CNS-effects were still present during acquisition of the PET scan. These findings suggest that it is unlikely that striatal dopamine release immediately after THC administration has been much larger than the modest levels of 3.4–3.9% that were observed around 45 min after administration. Indeed, effects on VAS 'feeling high' that were reported 101 min after inhalation were still significant.

Moreover, it is known that drug-induced effects on dopamine D_2/D_3 receptor availability last longer than changes in synaptic dopamine concentrations (Breier *et al*, 1997; Laruelle *et al*, 1997). In humans, [^{11}C]raclopride

BP was still decreased 6 h after amphetamine administration (Cardenas *et al*, 2004). This is probably because of an internalization of dopamine receptors (Laruelle, 2000), indicating that a drug-induced effect on striatal dopamine release can be detected for a long time after administration.

Our finding of THC-induced release of dopamine in the striatum suggests that human striatal dopamine release is under control of the endogenous cannabinoid system. The exact mechanism is still unclear, but cannabinoid CB1 receptors on glutamate and GABA terminals in both the nucleus accumbens and the ventral tegmental area are involved in the regulation of dopamine release in the striatum (Schlicker and Kathmann, 2001; Lupica *et al*, 2004). Interestingly, it is known that cannabis use increases the risk for developing schizophrenia (Arseneault *et al*, 2004; Moore *et al*, 2007) and worsens its clinical outcome (Linszen *et al*, 1994; D'Souza *et al*, 2005). Schizophrenia is an illness that has consistently been related to increased dopamine function in the striatum (Seeman and Lee, 1975; Angrist and Van Kammen, 1984), possibly caused by disinhibition of striatal dopamine transmission (Laruelle *et al*, 1996; Breier *et al*, 1997). Thus, elevated striatal dopamine release after the use of cannabis may explain how cannabis use contributes to the development and pathophysiology of schizophrenia.

In conclusion, we have demonstrated that Δ9-THC, the main psychoactive component of cannabis, induces dopamine release in the human striatum. This finding implies that THC may share a putatively addictive property with other drugs of abuse and that the endogenous cannabinoid system is involved in regulating striatal dopamine release, possibly explaining some of the detrimental effects of THC in neuropsychiatric disorders such as schizophrenia.

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DISCLOSURE/CONFLICT OF INTEREST

The authors declare that no financial support or compensation has been received from any individual or corporate entity over the past 3 years for research or professional service and there are no personal financial holdings that could be perceived as constituting a potential conflict of interest.

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