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Original Paper

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Abstract

Cannabis use in adolescence is emerging as a risk factor for the development of psychosis. In animal studies, Δ9-tetrahydrocannabinol (THC), the psychoactive component of cannabis, modulates striatal dopaminergic neurotransmission. Alterations in human striatal dopaminergic function have also been reported both in psychosis and in stimulant use. We sought to examine whether striatal dopamine D_2/D_3 receptor availability was altered in volunteers with a history of cannabis use using a database of previously acquired [^{11}C]-raclopride positron emission tomography (PET) scans. Ten [^{11}C]-raclopride scans from volunteers with a history of cannabis use were compared to ten control scans using a functional striatal subdivision region of interest (ROI) analysis. No significant differences in either overall striatal BP_{ND} values or BP_{ND} values in any functional striatal subdivision were found between the two groups. There was also no correlation between lifetime frequency of cannabis use and BP_{ND} values. Limbic striatal BP_{ND} values were ten percent lower in current nicotine cigarette smokers. These findings suggest that, unlike other drugs of abuse, a history of cannabis use is not associated with alterations in striatal dopamine D_2/D_3 receptor availability.

Keywords

[^{11}C]-raclopride, addiction, cannabis, D_2/D_3 receptor, dopamine, nicotine, PET, psychosis

Introduction

Cannabis (*Cannabis sativa*) is one of the most widely used recreational drugs in the world (Chawla, 2006). The psychoactive effects of cannabis are produced by Δ9-tetrahydrocannabinol (THC), which binds at cerebral cannabinoid CB_1 receptors (Ameri, 1999). Both CB_1 receptors and dopamine D_2 receptors are co-localized in the striatum (Pickel et al., 2006) and several studies demonstrate a modulatory effect of cannabinoids on dopaminergic neurotransmission. For example, in experimental animals, activation of CB_1 receptors produces inhibition of D_2 receptor-mediated responses (Marcellino et al., 2008) and THC administration increases both ventral striatal dopamine release (Cheer et al., 2004) and the firing rate of mesolimbic dopaminergic neurons (Tanda et al., 1997), although inhibition of dopamine release has also been reported (Sidlo et al., 2008).

In humans, the use of stimulants such as methamphetamine and cocaine, which increase striatal synaptic dopamine levels, is associated with decreased striatal dopamine D_2/D_3 receptor availability (Volkow et al., 1990, 2001). This may occur as a result of downregulation of postsynaptic D_2/D_3 receptors secondary to increases in synaptic dopamine levels produced by drug exposure, or alternatively could precede drug use, predisposing individuals to subsequent addiction (Volkow et al., 1999). Indeed, Volkow and colleagues suggest that high striatal dopamine D_2 receptor levels could protect against subsequent recreational drug use (Volkow et al., 2009). Furthermore, in animals, repeated exposure to CB_1 agonists such as THC leads to increases in the magnitude of responses to other drugs of abuse (Ferrari et al., 1999; Lamarque et al., 2001; Muschamp and Siviy, 2002), suggesting that some form of neurobiological sensitization, most likely in the dopaminergic system, occurs in response to cannabis exposure. However, it is currently unknown whether

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alterations in striatal dopamine D_2/D_3 receptor availability are associated with cannabis use.

Dopamine D_2/D_3 receptor availability in cannabis users is also of relevance to psychosis. Cannabis use in early adolescence is emerging as an environmental risk factor for the development of psychosis (Moore et al., 2007) and, in patients with schizophrenia, cannabis use may worsen the severity of positive psychotic symptoms and increase relapse rates (Grech et al., 2005; Linszen et al., 1994). Despite increasing evidence of a link between cannabis use and psychosis, the neurochemical process underlying this association is unclear. One possible mechanism is the modulation of striatal dopaminergic transmission by THC (Barkus et al., 2010; Bossong et al., 2009; Stokes et al., 2009) which may be reflected by alterations in striatal dopamine D_2/D_3 receptor availability.

In the present study, we examined striatal dopamine D_2/D_3 receptor availability in healthy volunteers with a history of cannabis use, using a database of previously acquired [^{11}C]-raclopride positron emission tomography (PET) scans. We hypothesized that a history of cannabis use would be associated with alterations in striatal dopamine D_2/D_3 receptor availability.

Methods

Participants and power analysis

Ten [^{11}C]-raclopride scans from volunteers with a history of cannabis use, defined as a lifetime history of using cannabis at least 50 times, were compared to ten scans from a control group consisting of volunteers who either had never used cannabis or whose past use was minimal (less than five times over their lifetime). The [^{11}C]-raclopride scans were obtained from four previous studies: three drug/cognitive challenge studies (Egerton et al., 2010; Lappin et al., 2009; Stokes et al., 2009) and one test–retest reliability study (Stokes et al., 2010). For the drug/cognitive challenge studies the placebo/resting state [^{11}C]-raclopride scan was included in the analysis and for the test–retest reliability the first scan of each pair was included. In total 6 resting state scans and 14 placebo scans were included in the analysis; five volunteers had previously undergone a THC challenge scan. The group size of ten volunteers was powered to detect a 9.5% change in overall [^{11}C]-raclopride binding, using a between-volunteer standard deviation (SD) of 7.2% for overall striatal [^{11}C]-raclopride binding previously reported by us (Stokes et al., 2009) and a power of 0.8 (power calculation generated using PS – http://biostat.mc.vanderbilt.edu/Power SampleSize).

Total lifetime cannabis, alcohol, nicotine and use of other substances were assessed for each volunteer either by the use of the cannabis experiences questionnaire (CEQ3) (Barkus et al., 2006) (35% of volunteers) or, if this was not available, by using volunteer self-reports of their use of alcohol, nicotine, cannabis and other substances provided as part of a screening questionnaire prior to imaging. All volunteers had been previously assessed by a psychiatrist to exclude current or previous significant mental health disorders and alcohol or recreational drug dependency as defined by the Diagnostic and Statistical Manual of Mental Disorders, fourth edition (DSM-IV), serious physical illness, past neurological disorders or previous use of psychotropic medications. All volunteers had previously given written informed consent for the study, which was approved by both the Hammersmith Research Ethics Committee and the Administration of Radioactive Substances Advisory Committee, UK.

Image acquisition and processing

On the day of imaging, all volunteers underwent urine drug screen analysis for THC, cocaine, methamphetamine, amphetamine, opiates and benzodiazepines. Any volunteer who produced a positive urine drug screen on the scan day was excluded from the study. Volunteers were also asked to abstain from alcohol for 24 hours prior to the scan day. All PET scans were acquired using an ECAT HR+ 962 scanner (CTI/Seimens) with an axial field of view of 15.5 cm. [^{11}C]-raclopride was administered as an initial intravenous bolus followed by constant infusion, with an infusion length of 70 minutes for scans from the Lappin et al. (2009) study (1 scan), 85 minutes for scans from the Stokes et al. (2009) (9 scans) and Egerton et al. (2010) (5 scans) studies, and 100 minutes for scans from the Stokes et al. (2010) study (5 scans). The bolus infusion approach mitigates blood flow changes affecting binding potential (BP_{ND}) values by establishing a state of constant equilibrium (Carson et al., 1997). A ten minute transmission scan was performed prior to each emission scan to measure and correct for tissue attenuation. Dynamic emission scans were acquired in three-dimensional mode using a standard acquisition protocol (22 time frames over 70 minutes for the Lappin et al. (2009) study, 28 time frames over 85 minutes for the Stokes et al. (2009) and Egerton et al. (2010) studies, and 38 frames over 100 minutes for the Stokes et al. (2010) study).

Image analysis

All dynamic scans were corrected for head movement using frame-by-frame (FBF) realignment (Montgomery et al., 2006). This procedure was applied to all frames to generate a FBF corrected dynamic image, which was then analysed using an automated region of interest (ROI) analysis. Striatal and cerebellar ROIs were defined using an atlas comprised of the three functional subdivisions of the striatum (limbic, associative and sensorimotor striatum) along with the cerebellum. These striatal subdivisions are anatomically analogous to the ventral striatum (limbic striatum), pre-commissural dorsal putamen, pre-commissural dorsal caudate and post-commissural dorsal caudate (associative striatum) and post-commissural putamen (sensorimotor striatum) (Martinez et al., 2003). An [^{11}C]-raclopride template was spatially transformed to the individual PET space of each FBF-corrected add image within SPM5 (www.fil.ion.ucl.ac.uk/spm) and the resulting deformation matrix was then applied to the atlas. A weighted steady-state average add image was then generated from each FBF-corrected dynamic image using in-house software written in Matlab (version 5; The MathWorks, Inc., Natick, Massachusetts, USA). The deformed striatal atlas was used to sample counts from the add image using Analyze 8.0 software (www.analyzedirect.com). BP_{ND} values, the ratio of specifically bound radioligand

Table 1. Volunteer demographics, alcohol, nicotine and cannabis use, and use of other substances

Variable	Volunteers with a history of cannabis use – mean (SD)	Control volunteers – mean (SD)	*p*-value
Age in years	32.6 (7.7)	36.5 (4.5)	0.19
Gender – % male	60	90	0.13
Alcohol consumption (units per week)	5.7 (4.6)	8.9 (5.9)	0.19
Current nicotine cigarette smokers (%)	30	20	0.61
Age of first cannabis use (years)	19.4 (3.5)	N/A	
Years of cannabis use (years)	11.8 (6.9)	N/A	
Total lifetime cannabis use (spliffs consumed)	2032 (3650)	0.1 (0.3)	0.001
Frequency of cannabis use per lifetime year (spliffs consumed)	52.3 (82.4)	N/A	
Last cannabis use (months)	17.8 (14.9)	N/A	
Total lifetime MDMA use (number of drug use sessions)	12.3 (35)	0	0.01
Total lifetime cocaine use (number of drug use sessions)	2.4 (3.3)	0	0.03
Total lifetime amphetamine use (number of drug use sessions)	7 (16.5)	0	0.03
Total lifetime opiate use (number of drug use sessions)	0	0	

to that of the non-displaceable ligand in the cerebellar reference tissue (Innis et al., 2007), were calculated for each striatal region as the ratio of striatal counts to cerebellar counts, minus 1, over the steady-state time period. The steady-state time period was defined as commencing at 39 minutes post-injection and continuing until the end of the scan, based on Watabe and colleague's estimates (Watabe et al., 2000) of the optimal timing for the establishment of the steady state. Thus the steady-state period duration was 31 minutes for the Lappin et al. (2009) study scan, 46 minutes for the Stokes et al. (2009) and Egerton et al. (2010) study scans, and 61 minutes for the Stokes et al. (2010) study scans. Accounting for decay, all studies used the same bolus to infusion ratio. We previously reported [^{11}C]-raclopride equilibrium stability of ± 0.06% per minute over the equilibrium period using this bolus infusion protocol (Stokes et al., 2009)

Statistical analysis

All data were checked for normality of distribution. Group differences in regional BP_{ND} values were assessed using a multivariate analysis of variance (MANOVA) for normally distributed variables and Mann-Whitney U-tests for non-normally distributed variables. Correlations between continuous data were assessed using Pearson's product moment correlation coefficient and discontinuous data with Spearman's rank correlation coefficient. All statistical comparisons were performed using SPSS 17.0 (SPSS, Chicago, Illinois, USA), all values are expressed as mean (SD) and the threshold for two-tailed statistical significance was defined as $p < 0.05$.

Results

Volunteer demographics and substance use

Volunteer demographics, alcohol, nicotine and cannabis use and use of other substances are shown in Table 1. Volunteers who were current tobacco cigarette smokers last smoked at least three hours before imaging.

There was no significant difference in age, gender, current alcohol consumption or percentage of current nicotine cigarette smokers between volunteers with a history of cannabis use and controls (all *p*-values > 0.05). One volunteer from the control group used cannabis once at the age of 16 and none of the control group had ever used any other substances.

Volunteers with a history of cannabis use first used cannabis at the age of 19 (3.5) years (min. 16 years, max. 28 years). They had used cannabis an average of 2032 (3650) times over a period of 11.8 (6.9) years, with a mean frequency of cannabis cigarettes consumed per lifetime year of 52 (82). Volunteers with a history of cannabis use had last used cannabis on average 17.8 (14.9) months before imaging. They also had significantly higher total mean lifetime usage of cocaine (2.4 (3.3) times), amphetamine (7 (16.5) times) and MDMA (12.3 (35) times) than the control group (see Table 1).

Radiochemistry

There was no significant difference in the radioactivity dose injected or the amount of unlabelled raclopride co-injected between the two groups (*p*-values > 0.05).

Striatal D_2/D_3 receptor availability

Association with demographic, radiochemical and scan variables. Increasing age was significantly associated with reductions in total striatal BP_{ND} values ($F_{1,15}$=4.6, $p = 0.05$) and BP_{ND} values in the associative ($F_{1,15}$=4.5, $p = 0.05$) and sensorimotor ($F_{1,15}$= 4.3, $p = 0.05$) subdivisions, but not the limbic subdivision ($F_{1,15}$= 1.4, $p = 0.26$).

Gender had no effect on either total striatal BP_{ND} values or values in any striatal subdivision (all *p* values > 0.05). [^{11}C]-raclopride bolus-infusion duration length, number of PET scan acquisition frames, whether the scan was acquired after placebo administration, or whether the volunteer had previously undergone a THC challenge scan had no significant effect on either total striatal BP_{ND} values or BP_{ND} values in any functional subdivision (*p*-values > 0.05).

Table 2. Total striatal and functional striatal [^{11}C]-raclopride BP_{ND} values in volunteers with a history of cannabis use and control volunteers

Striatal area	Volunteers with a history of cannabis use BP_{ND} (SD)	Control volunteers BP_{ND} (SD)	*F* statistic	*p*-value
Total	2.53 (0.19)	2.45 (0.22)	0.86	0.37
Limbic	2.32 (0.29)	2.28 (0.20)	0.10	0.76
Associative	2.44 (0.18)	2.33 (0.24)	1.4	0.25
Sensorimotor	2.85 (0.25)	2.80 (0.23)	0.2	0.65

Association with alcohol use, nicotine use and substance use. Current alcohol consumption had no effect on either total striatal BP_{ND} values or BP_{ND} values from any functional subdivision (all p-values > 0.05). Limbic striatal BP_{ND} values were significantly lower in current nicotine smokers ($F_{1,15} = 4.34$, $p = 0.05$) with five current nicotine users having 10% lower BP_{ND} values than the 15 volunteers imaged who did not currently smoke nicotine cigarettes (current nicotine smokers limbic BP_{ND}: 2.12 (0.22), non-nicotine smokers limbic BP_{ND}: 2.36 (0.22)). Age did not significantly differ between current nicotine smokers and non-smokers (age of current nicotine smokers: 36.0 (6.7), age of non smokers: 34.1 (6.6), $p = 0.57$). There was no significant association between current nicotine use and either total striatal BP_{ND} values or BP_{ND} values in any other functional subdivision (p-values > 0.05). A history of any previous stimulant (amphetamine or cocaine) use or any previous MDMA use had no significant effect on either total striatal BP_{ND} values or BP_{ND} values in any other functional subdivision (p-values > 0.05)

Association with a history of cannabis use. There was no significant difference in [^{11}C]-raclopride BP_{ND} values between volunteers with a history of cannabis use and controls for either the total striatum or any of the three functional striatal subdivisions (all p-values > 0.05) (Table 2). These results did not change if volunteer age or current nicotine cigarette smoking status were included as covariates in the analysis. Total striatal [^{11}C]-raclopride BP_{ND} values for volunteers with a history of cannabis use compared to controls are shown in Figure 1.

There was no correlation between the frequency of lifetime cannabis use, years of cannabis use, age of first use or duration since last cannabis use and BP_{ND} values in either the total striatum or any functional subdivision (all p-values > 0.05).

Discussion

We found that a history of cannabis use was not associated with alterations in dopamine D_2/D_3 receptor availability either in the whole striatum or any of the striatal functional subdivisions. We also found no correlation between the frequency of lifetime cannabis use and D_2/D_3 receptor availability, even though volunteers with a history of cannabis use had used cannabis on average over two thousand times over their lifetime. These results are consistent with experimental animal studies which report no alterations in striatal dopamine D_2 receptors after repeated cannabinoid exposure (Dalton and Zavitsanou, 2010; Higuera-Matas et al., 2010) and no alterations in striatal D_2 receptor expression in transgenic mice deficient in CB_1 receptors (Gerald et al., 2006). They also concur with a human pilot study where no differences in striatal D_2/D_3 receptor availability were found in a cohort of six cannabis dependent volunteers in early remission (Sevy et al., 2008).

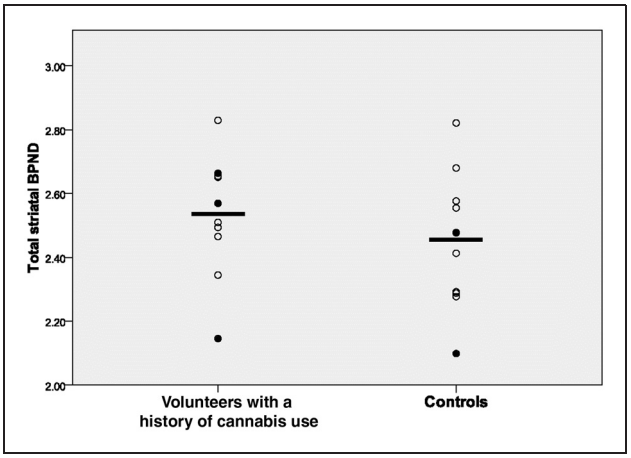

Figure 1. Total striatal [^{11}C]-raelopride BP_{ND} values for volunteers with a history of cannabis use compared to control volunteers. (Bar indicates mean group value, filled circles indicate current tobacco smokers).

Our findings indicate that cannabis use is not associated with alterations in human striatal D_2/D_3 receptor availability. Therefore, if cannabis use is associated with an increased risk for further recreational use of other drugs, there is no indication from our results that this is mediated by changes in striatal D_2/D_3 receptor availability. We would suggest that if these associations are correct then they may be mediated by other neurochemical mechanisms such alterations in CB_1 receptors or dopamine D_1 receptor availability. These results also do not support theories that decreased striatal D_2/D_3 receptor availability predisposes volunteers to use cannabis or that cannabis use could lead to downregulation of the striatal dopaminergic system as a consequence of dopamine release. Indeed, recent human neurochemical imaging studies provide inconclusive evidence that THC modulates striatal dopamine release. Stokes et al. (2009) found no striatal dopamine release after an oral THC challenge. In a smaller cohort of volunteers, Bossong and colleagues found a modest decrease in striatal [^{11}C]-raclopride binding which fell within test re-test imaging reliability after an inhaled THC challenge (Bossong et al., 2009) and Barkus and colleagues found no striatal dopamine release after an intravenous THC challenge, despite inducing psychotic symptoms in volunteers (Barkus et al., 2010).

A caveat to the interpretation of our results is that we cannot exclude the possibility that, with abstinence, the striatal D_2/D_3 receptor system may have recovered from the effects of cannabis use. Nader and colleagues found in a primate study that the D_2/D_3 receptor system can recover over a period of a year after chronic cocaine use in some, but by no

means all, individuals (Nader et al., 2006). In our study, volunteers with a history of cannabis use were imaged on average 18 months after their last use of cannabis. However, we found no correlation between duration of last cannabis use and BP_{ND} values in either the overall striatum or any functional striatal subdivision. Moreover the Sevy et al. (2008) study also found no alterations in striatal D_2/D_3 receptor availability in a cohort of volunteers who had last used cannabis an average of fifteen weeks before imaging.

In addition to determining whether a history of cannabis use was associated with alterations in striatal D_2/D_3 receptor availability, we also found that volunteers who were current nicotine cigarette smokers had significantly decreased limbic D_2/D_3 receptor availability. Although the 10% reduction in limbic D_2/D_3 receptor availability found in current nicotine cigarette users is a preliminary finding from a small cohort of volunteers ($n = 5$), this result concurs with previous studies demonstrating reductions in limbic striatal D_2/D_3 receptor availability after a nicotine smoking session indicative of dopamine release (Brody et al., 2004, 2009). It is also in agreement with a study of current nicotine cigarette smokers which reported reductions in striatal D_2/D_3 receptor availability; although in this study the reductions were found in the putamen rather than the limbic striatum (Fehr et al., 2008). As alterations in limbic striatal dopaminergic function have been postulated to be critical to the underlying neurobiology of nicotine addiction (Pierce and Kumaresan, 2006), we would suggest that this potentially important finding is further explored in a larger cohort of current nicotine cigarette smokers.

There are a number of potential limitations to this study. First, we based our analysis on volunteer self-reports of their lifetime use of cannabis and other substances. As these reports are based on subjective recall, the possibility remains that volunteers may have either over- or underestimated their use of recreational drugs, which could have potentially affected our results. Second, it is possible that volunteers with a history of cannabis use who were imaged in this study may not have used cannabis heavily enough to affect the striatal dopaminergic system. Although the definition of 'heavy' cannabis use is necessarily arbitrary, Solowij and colleagues used criteria of heavy chronic cannabis use, which included regular use more than once a week, for more than three years (Solowij et al., 2002). As 7 out of 10 volunteers with a history of cannabis use included in this study would have fulfilled these criteria, we would contend that our study reflected striatal D_2/D_3 receptor availability in volunteers with a history of cannabis use in the medium to heavy end of the cannabis-use spectrum. Third, although we did not find a placebo effect on [^{11}C]-raclopride binding, it may still be possible that the placebo condition affected binding. Several studies have reported alterations in [^{11}C]-raclopride binding after receiving a placebo associated with the expectation of a reward, such as an improvement in symptoms in Parkinson's disease or relief from pain (de la Fuente-Fernandez et al., 2001; Lidstone et al., 2010; Zubieta and Stohler, 2009). Finally, we cannot exclude the possibility that small differences in D_2/D_3 receptor availability may exist between volunteers with a history of cannabis use and controls. Our study was powered to reliably detect a 9.5% difference in overall [^{11}C]-raclopride binding which is adequately powered to detect, for example, a 15% decrease in overall [^{11}C]-raclopride binding previously reported in chronic cocaine users (Martinez et al., 2004). However, to reliably detect smaller group differences of five percent or less would require a considerably larger study including at least 35 volunteers per group.

In summary, we have shown that a history of cannabis use is not associated with alterations in striatal D_2/D_3 receptor availability. Our findings add to the literature questioning the effects of cannabis use on the human striatal dopamine D_2/D_3 receptor system. We would suggest that further studies of the neurochemical effects of cannabis use focus on other neuroreceptor systems such as the cannabinoid CB1 receptor or the dopamine D_1 receptor.

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References

Ameri A (1999) The effects of cannabinoids on the brain. *Prog Neurobiol* 58: 315–348.

Barkus E, Morrison PD, Vuletic D, Dickson J, Ell PJ, Pilowsky LS, et al. (2010) Does intravenous Δ9-tetrahydrocannabinol increase dopamine release? A SPET study. *J Psychopharmacol* 25: 1462–1468.

Barkus EJ, Stirling J, Hopkins RS and Lewis S (2006) Cannabis-induced psychosis-like experiences are associated with high schizotypy. *Psychopathology* 39: 175–178.

Bossong MG, van Berckel BN, Boellaard R, Zuurman L, Schuit RC, Windhorst AD, et al. (2009) Delta 9-tetrahydrocannabinol induces dopamine release in the human striatum. *Neuropsychopharmacology* 34: 759–766.

Brody AL, Mandelkern MA, Olmstead RE, Allen-Martinez Z, Scheibal D, Abrams AL, et al. (2009) Ventral striatal dopamine release in response to smoking a regular vs a denicotinized cigarette. *Neuropsychopharmacology* 34: 282–289.

Brody AL, Olmstead RE, London ED, Farahi J, Meyer JH, Grossman P, et al. (2004) Smoking-induced ventral striatum dopamine release. *Am J Psychiatry* 161: 1211–1218.

Carson RE, Breier A, de Bartolomeis A, Saunders RC, Su TP, Schmall B, et al. (1997) Quantification of amphetamine-induced changes in [^{11}C]raclopride binding with continuous infusion. *J Cereb Blood Flow Metab* 17: 437–447.

Chawla S (2006) *World Drug Report 2006*. New York City, NY: United Nations Office on Drugs and Crime.

Cheer JF, Wassum KM, Heien ML, Phillips PE and Wightman RM (2004) Cannabinoids enhance subsecond dopamine release in the nucleus accumbens of awake rats. *J Neurosci* 24: 4393–4400.

Dalton VS and Zavitsanou K (2010) Differential treatment regimen-related effects of cannabinoids on D1 and D2 receptors in adolescent and adult rat brain. *J Chem Neuroanat* 40: 272–280.

de la Fuente-Fernandez R, Ruth TJ, Sossi V, Schulzer M, Calne DB and Stoessl AJ (2001) Expectation and dopamine release: mechanism of the placebo effect in Parkinson's disease. *Science* 293: 1164–1166.

Egerton A, Shotbolt JP, Stokes PR, Hirani E, Ahmad R, Lappin JM, et al. (2010) Acute effect of the anti-addiction drug bupropion on extracellular dopamine concentrations in the human striatum: An [^{11}C]raclopride PET study. *Neuroimage* 50: 260–266.

Fehr C, Yakushev I, Hohmann N, Buchholz HG, Landvogt C, Deckers H, et al. (2008) Association of low striatal dopamine

d2 receptor availability with nicotine dependence similar to that seen with other drugs of abuse. *Am J Psychiatry* 165: 507–514.

Ferrari F, Ottani A and Giuliani D (1999) Influence of the cannabinoid agonist HU 210 on cocaine- and CQP 201-403-induced behavioural effects in rat. *Life Sci* 65: 823–831.

Gerald TM, Ward GR, Howlett AC and Franklin SO (2006) CB1 knockout mice display significant changes in striatal opioid peptide and D4 dopamine receptor gene expression. *Brain Res* 1093: 20–24.

Grech A, Van Os J, Jones PB, Lewis SW and Murray RM (2005) Cannabis use and outcome of recent onset psychosis. *Eur Psychiatry* 20: 349–353.

Higuera-Matas A, Botreau F, Del Olmo N, Miguens M, Olias O, Montoya GL, et al. (2010) Periadolescent exposure to cannabinoids alters the striatal and hippocampal dopaminergic system in the adult rat brain. *Eur Neuropsychopharmacol* 20: 895–906.

Innis RB, Cunningham VJ, Delforge J, Fujita M, Gjedde A, Gunn RN, et al. (2007) Consensus nomenclature for in vivo imaging of reversibly binding radioligands. *J Cereb Blood Flow Metab* 27: 1533–1539.

Lamarque S, Taghzouti K and Simon H (2001) Chronic treatment with Delta(9)-tetrahydrocannabinol enhances the locomotor response to amphetamine and heroin. Implications for vulnerability to drug addiction. *Neuropharmacology* 41: 118–129.

Lappin JM, Reeves SJ, Mehta MA, Egerton A, Coulson M and Grasby PM (2009) Dopamine release in the human striatum: motor and cognitive tasks revisited. *J Cereb Blood Flow Metab* 29: 554–564.

Lidstone SC, Schulzer M, Dinelle K, Mak E, Sossi V, Ruth TJ, et al. (2010) Effects of expectation on placebo-induced dopamine release in Parkinson disease. *Arch Gen Psychiatry* 67: 857–865.

Linszen DH, Dingemans PM and Lenior ME (1994) Cannabis abuse and the course of recent-onset schizophrenic disorders. *Arch Gen Psychiatry* 51: 273–279.

Marcellino D, Carriba P, Filip M, Borgkvist A, Frankowska M, Bellido I, et al. (2008) Antagonistic cannabinoid CB1/dopamine D2 receptor interactions in striatal CB1/D2 heteromers. A combined neurochemical and behavioral analysis. *Neuropharmacology* 54: 815–823.

Martinez D, Slifstein M, Broft A, Mawlawi O, Hwang DR, Huang Y, et al. (2003) Imaging human mesolimbic dopamine transmission with positron emission tomography. Part II: Amphetamine-induced dopamine release in the functional subdivisions of the striatum. *J Cereb Blood Flow Metab* 23: 285–300.

Martinez D, Broft A, Foltin RW, Slifstein M, Hwang DR, Huang Y, et al. (2004) Cocaine dependence and d2 receptor availability in the functional subdivisions of the striatum: relationship with cocaine-seeking behavior. *Neuropsychopharmacology* 29: 1190–1202.

Montgomery AJ, Thielemans K, Mehta MA, Turkheimer F, Mustafovic S and Grasby PM (2006) Correction of head movement on PET studies: comparison of methods. *J Nucl Med* 47: 1936–1944.

Moore TH, Zammit S, Lingford-Hughes A, Barnes TR, Jones PB, Burke M, et al. (2007) Cannabis use and risk of psychotic or affective mental health outcomes: a systematic review. *Lancet* 370: 319–328.

Muschamp JW and Siviy SM (2002) Behavioral sensitization to amphetamine follows chronic administration of the CB1 agonist WIN 55,212-2 in Lewis rats. *Pharmacol Biochem Behav* 73: 835–842.

Nader MA, Morgan D, Gage HD, Nader SH, Calhoun TL, Buchheimer N, et al. (2006) PET imaging of dopamine D2 receptors during chronic cocaine self-administration in monkeys. *Nat Neurosci* 9: 1050–1056.

Pickel VM, Chan J, Kearn CS and Mackie K (2006) Targeting dopamine D2 and cannabinoid-1 (CB1) receptors in rat nucleus accumbens. *J Comp Neurol* 495: 299–313.

Pierce RC and Kumaresan V (2006) The mesolimbic dopamine system: the final common pathway for the reinforcing effect of drugs of abuse? *Neurosci Biobehav Rev* 30: 215–238.

Sevy S, Smith GS, Ma Y, Dhawan V, Chaly T, Kingsley PB, et al. (2008) Cerebral glucose metabolism and D2/D3 receptor availability in young adults with cannabis dependence measured with positron emission tomography. *Psychopharmacology (Berl)* 197: 549–556.

Sidlo Z, Reggio PH and Rice ME (2008) Inhibition of striatal dopamine release by CB1 receptor activation requires nonsynaptic communication involving GABA, H_2O_2, and KATP channels. *Neurochem Int* 52: 80–88.

Solowij N, Stephens RS, Roffman RA, Babor T, Kadden R, Miller M, et al. (2002) Cognitive functioning of long-term heavy cannabis users seeking treatment. *JAMA* 287: 1123–1131.

Stokes PR, Egerton A, Watson B, Reid A, Breen G, Lingford-Hughes A, et al. (2010) Significant decreases in frontal and temporal [^{11}C]-raclopride binding after THC challenge. *Neuroimage* 52: 1521–1527.

Stokes PR, Mehta MA, Curran HV, Breen G and Grasby PM (2009) Can recreational doses of THC produce significant dopamine release in the human striatum? *Neuroimage* 48: 186–190.

Tanda G, Pontieri FE and Di Chiara G (1997) Cannabinoid and heroin activation of mesolimbic dopamine transmission by a common mu1 opioid receptor mechanism. *Science* 276: 2048–2050.

Volkow ND, Chang L, Wang GJ, Fowler JS, Ding YS, Sedler M, et al. (2001) Low level of brain dopamine D2 receptors in methamphetamine abusers: association with metabolism in the orbitofrontal cortex. *Am J Psychiatry* 158: 2015–2021.

Volkow ND, Fowler JS, Wang GJ, Baler R and Telang F (2009) Imaging dopamine's role in drug abuse and addiction. *Neuropharmacology* 56(Suppl 1): 3–8.

Volkow ND, Fowler JS, Wolf AP, Schlyer D, Shiue CY, Alpert R, et al. (1990) Effects of chronic cocaine abuse on postsynaptic dopamine receptors. *Am J Psychiatry* 147: 719–724.

Volkow ND, Wang GJ, Fowler JS, Logan J, Gatley SJ, Gifford A, et al. (1999) Prediction of reinforcing responses to psychostimulants in humans by brain dopamine D2 receptor levels. *Am J Psychiatry* 156: 1440–1443.

Watabe H, Endres CJ, Breier A, Schmall B, Eckelman WC and Carson RE (2000) Measurement of dopamine release with continuous infusion of [^{11}C]raclopride: optimization and signal-to-noise considerations. *J Nucl Med* 41: 522–530.

Zubieta JK and Stohler CS (2009) Neurobiological mechanisms of placebo responses. *Ann NY Acad Sci* 1156: 198–210.