BJPsych | The British Journal of Psychiatry (2011) 199, 391–397. doi: 10.1192/bjp.bp.110.084178

Effects of lowered serotonin transmission on cocaine-induced striatal dopamine response: PET [^{11}C]raclopride study in humans†

Sylvia M. L. Cox, Chawki Benkelfat, Alain Dagher, J. Scott Delaney, France Durand, Theodore Kolivakis, Kevin F. Casey and Marco Leyton

Background
Low serotonin transmission is thought to increase susceptibility to a wide range of substance use disorders and impulsive traits.

Aims
To investigate the effects of lowered serotonin on cocaine-induced (1.0 mg/kg cocaine, self-administered intranasally) dopamine responses and drug craving.

Method
In non-dependent cocaine users, serotonin transmission was reduced using the acute tryptophan depletion method. Striatal dopamine responses were measured using positron emission tomography with [^{11}C]raclopride.

Results
Acute tryptophan depletion increased drug craving and striatal dopamine responses to cocaine. These acute tryptophan depletion-induced increases did not occur in the absence of cocaine.

Conclusions
The results suggest that low serotonin transmission can increase dopaminergic and appetitive responses to cocaine. These findings might identify a mechanism by which individuals with low serotonin are at elevated risk for both substance use disorders and comorbid conditions.

Declaration of interest
None.

Low serotonin (5-HT) transmission is thought to increase susceptibility to a wide range of substance use disorders and impulsive behaviours.[1–3] In part, this might reflect an influence of 5-HT on ascending midbrain dopamine pathways. For example, in laboratory animals 5-HT can alter dopamine cell firing and release.[4–6] Moreover, increasing and decreasing 5-HT transmission can diminish and augment, respectively, the ability of drugs such as cocaine to induce a dopamine response.[7–9] Decreasing 5-HT transmission can also augment cocaine's behavioural effects, enhancing self-administration responses and increasing cocaine-induced locomotor activity, conditioned place preferences and conditioned approach.[7,10,11] Increasing 5-HT transmission, in comparison, diminishes these effects.[8,9,12] In humans, the effect of serotonergic manipulations on drug-induced dopamine responses has not been investigated. To examine these hypothesised interactions, the present study tested the effect of an experimentally induced low 5-HT state on the rewarding and dopaminergic effects of cocaine. Central 5-HT transmission was reduced using the acute tryptophan depletion method. Dopamine responses were measured using positron emission tomography (PET) plus the labelled tracer [^{11}C]raclopride. It was predicted that the induction of a low 5-HT state would increase both dopamine and appetitive responses to cocaine.

Method

The studies were carried out in accordance with the Declaration of Helsinki and were approved by the Research Ethics Board of the Montreal Neurological Institute. All participants gave written informed consent.

Participants

Seventeen non-dependent cocaine users (2 females, mean age 24.8 years, s.d. = 3.7) (Table 1) were recruited through advertisements in local newspapers. Ten of these participants are the same individuals as those in our previous report.[13] For all participants, the primary and preferred route of self-administration was intranasal. All were free of current or past substance dependence, as determined by a semi-structured clinical interview for DSM–IV diagnoses.[14] In total 9 of the 17 participants were current smokers with a score of four or less on the Fagerström Test for Nicotine Dependence (FTND)[15] (mean 0.6, s.d. = 1.3, $n = 9$). All were free of current substance misuse. Women were tested during the follicular phase.

Participants were free of any other current Axis I psychopathology and were physically healthy as determined by physical examination, electrocardiography and standard laboratory tests. Prior to each test session, participants abstained from alcohol for at least 24 h and from nicotine for at least 12 h, with the exception of one participant in experiment two (with an FTND score of 4) who smoked one cigarette on the morning of each test day (about 6 h prior to each PET scan) in order to minimise nicotine withdrawal effects. On the morning of each test day, all tested negative on a urine drug screen sensitive to cocaine, opiates, phencyclidine, barbiturates, Δ^9–tetrahydrocannabinol, benzodiazepines and amphetamines (Triage Panel for Drugs of Abuse, Biosite Diagnostics, San Diego, California, USA, www.biosite.com).

Procedure

Experiment 1

All participants underwent PET [^{11}C]raclopride scans following the self-administration of:

(a) condition 1: cocaine hydrochloride (1.0 mg/kg) intranasally plus a tryptophan-deficient amino acid mixture (acute tryptophan depletion) orally;

(b) condition 2: cocaine (1.0 mg/kg) plus a nutritionally balanced amino acid mixture; and

†See editorial, pp. 353–354, this issue.

Table 1 Drug-use histories

	Experiment 1 ($n = 10$)		Experiment 2 ($n = 7$)	
	Participants exposed to drug, *n*	Mean (s.d.) total range	Participants exposed to drug, *n*	Mean (s.d.) total range
Cocaine				
Occasions used, past 12 months	10	18.4 (6.0) 12–28	7	12.3 (8.7) 6–30
Occasions used, lifetime	10	43.9 (25.9) 18–100	7	72.1 (66.4) 18–200
Quantity used per occasion in past 12 months, g	10	0.38 (0.25) 0.125–1	7	0.29 (0.12) 0.2–0.5
Amphetamine, occasions used, lifetime	6	18.9 (16.4) 0–45	4	26.8 (28.1) 0–60
MDMA, occasions used, lifetime	9	20.8 (22.6) 0–62	7	18.7 (13.4) 2–40
Ephedrine, occasions used, lifetime	3	21.7 (17.6) 0–40	0	
Opiates, occasions used, lifetime	1	60	2	1 (0) 0–1
Cannabis, occasions used, lifetime	10	388 (610) 10–1550[a]	7	565.7 (588.6) 20–1500
Psilocybin, occasion used, lifetime	8	14 (12.5) 0–35	6	3.5 (2.1) 0–6
LSD, occasions used, lifetime	7	14.1 (16.2) 0–50	3	5.3 (2.5) 0–8
Ketamine, occasions used, lifetime	5	5.8 (6.4) 0–15	0	
Alcohol, occasions used, lifetime	10	394 (279) 100–816[b]	7	632.1 (355.4) 175–1270

MDMA, 3,4-Methylenedioxymethamphetamine; LSD, lysergic acid diethylamide.
a. Missing data on four participants for lifetime use of cannabis.
b. Missing data on five participants for lifetime use of alcohol.

(c) condition 3: placebo powder (100 mg lactose) plus nutritionally balanced amino acid mixture.

The effect of cocaine alone (condition 2 *v.* 3) has recently been described elsewhere.[13] Here we focus on the effect of acute tryptophan depletion on cocaine-induced dopamine release. In order to keep the physical amount of cocaine and placebo powder constant, the active drug was mixed with lactose to yield a total of 100 mg. All test sessions were carried out under double-masked conditions, in a randomised, counterbalanced order.

On each test day, participants came to the laboratory at 09.00 h, were weighed and ingested an amino acid mixture, as described previously.[16] Then 4.5 h post-amino acid mixture, participants self-administered cocaine or placebo and were scanned immediately afterward. Self-report questionnaires, cardiovascular measures and blood tests to assess plasma amino acid levels were administered at five time points:

(a) baseline, prior to amino acid drink;

(b) 4.5 h post-amino acid drink, immediately prior to cocaine self-administration;

(c) immediately after tracer injection, about 15 min post-cocaine self-administration;

(d) mid-scan, 45 min post-cocaine self-administration; and

(e) the end of the scan, 75 min post-cocaine self-administration.

Cocaine plasma concentrations were obtained at times (b) to (e). Throughout testing, a project-dedicated nurse and emergency medicine physician were onsite to monitor participant's safety and a psychiatrist was on-call. Following each test session participants remained overnight for observation. Prior to being discharged, participants were evaluated by an emergency medicine physician. There were no serious adverse events.

Experiment 2

A second study was conducted to assess the effect of acute tryptophan depletion on [^{11}C]raclopride binding values in the absence of cocaine self-administration. Participants underwent PET scans with the labelled tracer [^{11}C]raclopride following the ingestion of:

(a) a tryptophan-deficient amino acid mixture (acute tryptophan depletion); and

(b) a nutritionally balanced amino acid mixture.

Both sessions were carried out under double-blind conditions, in a randomised, counterbalanced order. On each test day, participants ingested an amino acid mixture and were scanned 4.5 h later. Blood draws to assess plasma amino acid levels were collected at three different time points:

(a) baseline, prior to amino acid drink;

(b) 4.5 h post-amino acid drink; and

(c) at the end of the scan.

Image acquisition and processing

All participants were scanned on a Siemens ECAT HR + PET scanner (CTI/Siemens, Knoxville, Tenn) with lead septa removed (63-slice coverage, with a maximum resolution of 4.2 mm full-width at half maximum (FWHM) in the centre of the field of view (FOV)). Immediately after the transmission scan 7 mCi of [^{11}C]raclopride was injected as a bolus into the antecubital vein. Emission data were collected over 60 min in 26 time frames of progressively longer duration.

For anatomical coregistration, high-resolution (1 mm) T_1-weighted magnetic resonance images (MRI) were obtained for all participants on a 1.5 T Siemens scanner, using gradient echo pulse sequence (repetition time (TR) = 9.7 ms, echo time (TE) = 4 ms, flip angle 12°, FOV = 250 and matrix 256 × 256). These volumes were corrected for image intensity non-uniformity,[17] and linearly and non-linearly transformed into standardised stereotaxic Talairach-like space[18] using automated feature matching to the MNI305 template.[19] Each individual's MRI was then coregistered to their summed radioactivity PET images.[20]

The PET images were reconstructed using a 6 mm FWHM Hanning filter and corrected for movement.[21] Parametric images were generated by computing [^{11}C]raclopride binding potential (BP_{ND}) at each voxel using a simplified kinetic model that uses the cerebellum as a reference tissue devoid of dopamine $D_{2/3}$ receptors to describe the kinetics of the free and specifically bound ligand.[22] The BP_{ND} expresses the relationship between the estimated concentration of available dopamine D_2/D_3 receptors (B_{avail}), the dissociation constant of the radiotracer from

https://doi.org/10.1192/bjp.bp.110.084178 Published online by Cambridge University Press

D_2/D_3 receptors (K_d) and the free fraction of non-specifically bound tracer in the brain (F_{ND}):

$$BP_{ND} = F_{ND} \times (B_{avail}/K_D)$$

In a resting condition BP_{ND} is proportional to the concentration of available D_2/D_3 receptors; BP_{ND} decreases when dopamine release is elicited, and the magnitude of the change in BP_{ND} has been shown to be proportional to the increase in dopamine transmission.[23] Next, *t*-maps were generated, representing *t*-tests of voxel-by-voxel changes in [^{11}C]raclopride BP_{ND}[24] between the following conditions.

Experiment 1

(a) Nutritionally balanced amino acid mixture + cocaine *v.* nutritionally balanced amino acid mixture + placebo;

(b) acute tryptophan depletion + cocaine *v.* nutritionally balanced amino acid mixture + cocaine;

(c) acute tryptophan depletion + cocaine *v.* nutritionally balanced amino acid mixture + placebo.

Experiment 2

(a) Acute tryptophan depletion *v.* nutritionally balanced amino acid mixture.

For both experiments, voxels of statistically significant changes were identified by thresholding the *t*-maps at a value of $t \geqslant 4.1$ (adjusted for sample size, Experiment 1: $t = 4.05$, Experiment 2: $t \geqslant 4.12$) which corresponds to $P \leqslant 0.05$ Bonferroni corrected for multiple comparisons, based on a search volume of the entire striatum and a spatial resolution of 8 mm at FWHM.[25] To exclude potential noise, clusters of less than 5 voxels were ignored.

Self-report scales

Drug craving, mood and other subjective states were assessed with 14 visual analogue scale (VAS) items (happy, rush, high, euphoria, excited, anxious, energetic, mind-racing, alert, like drug effect, want cocaine, desire cocaine, urge for cocaine and crave cocaine) anchored at 1 (least) and 10 (most). A total craving score was calculated by combining the scores on all four craving-related VAS items: 'want cocaine', 'urge for cocaine', 'desire cocaine' and 'crave cocaine'.

Plasma amino acid and cocaine levels

Tryptophan and tyrosine levels were measured using gradient reverse-phase high-performance liquid chromatography with fluorometric detection. In Experiment 1, tryptophan levels were available from six participants at five time points whereas tyrosine levels were available for eight participants at three time points: baseline, prior to amino acid drink; 4.5 h post-amino acid drink; and at the end of the scan. In Experiment 1, plasma levels of cocaine were analysed with gas chromatography mass spectroscopy, using solid phase extraction. Data were missing from two participants ($n = 8$). For Experiment 2, tryptophan levels were missing from one participant ($n = 6$); tyrosine levels were missing from two participants ($n = 5$).

Statistics

All data were analysed using repeated measures ANOVAs, followed by Greenhouse–Geisser corrections and planned comparisons as appropriate (SPSS version 12).

Results

Plasma levels of tryptophan, tyrosine and cocaine

As expected, tryptophan values increased and decreased on the nutritionally balanced amino acid and acute tryptophan depletion tests, respectively, as reflected by test × time interactions (Experiment 1: $F(8, 40) = 25.3$, $P \leqslant 0.001$; Experiment 2: $F(2, 10) = 71.4$, $P \leqslant 0.0001$). Importantly, cocaine administration did not alter the magnitude of the tryptophan increase nor its time course on the sessions with the nutritionally balanced amino acid mixtures ($P = 0.4$). Ingestion of the acute tryptophan depletion mixture led to significant decreases in plasma tryptophan levels that were sustained throughout the scanning procedure (Experiment 1: $P \leqslant 0.0001$; Experiment 2: $P \leqslant 0.0001$) (Table 2). In comparison, acute tryptophan depletion did not affect plasma levels of tyrosine as indicated by the absence of test × time interactions (Experiment 1: $F(4, 28) = 1.17$, $P = 0.346$; Experiment 2: $F(2, 8) = 3.05$, $P = 0.104$).

In Experiment 1, plasma concentrations of cocaine differed significantly between the three test sessions as reflected by a test × time interaction ($F(6, 42) = 24.4$, P=0.0001). Plasma cocaine levels increased significantly in the cocaine conditions compared with nutritionally balanced amino acid mixture + placebo ($P = 0.0001$ for all time points). Plasma cocaine levels peaked at the mid-scan time point, 45 min post-cocaine self-administration (mean 149 ng/ml (s.d. = 44) and 155 ng/ml (s.d. = 32) for nutritionally balanced amino acid mixture + cocaine and acute tryptophan depletion + cocaine respectively); the plasma cocaine levels did not differ on the two drug test sessions ($P \geqslant 0.4$ for all time points). Cocaine was not detected in plasma on any of the drug-free test days or on any baseline measure prior to cocaine self-administration.

PET data: analyses of *t*-maps

Experiment 1

As previously reported,[13] cocaine plus nutritionally balanced amino acid mixture *v.* placebo powder plus nutritionally

Table 2 Plasma concentrations of tryptophan at five (Experiment 1) and three (Experiment 2) different time points

	Pre-amino acid		4.5 h		Start scan		Mid-scan		Post-scan	
Treatment	*n*	μg/ml, mean (s.d.)	*n*	μg/ml, mean (s.d.)	*n*	μg/ml, mean (s.d.)	*n*	μg/ml, mean (s.d.)	*n*	μg/ml, mean (s.d.)
Nutritionally balanced amino acid mixture + placebo	10	11.2 (1.3)	10	19.8 (6.1)	9	17.1 (5.3)	10	14.7 (4.5)	10	12.7 (4.0)
Nutritionally balanced amino acid mixture + cocaine	10	11.0 (1.5)	8	23.7 (5.9)	10	19.4 (5.2)	9	17.1 (5.0)	10	15.1 (5.1)
Acute tryptophan depletion + cocaine	10	11.9 (2.2)	10	1.5 (0.6)	10	1.2 (0.6)	9	1.2 (0.7)	10	1.4 (0.8)
Nutritionally balanced amino acid mixture	6	10.3 (0.6)	7	21.5 (5.3)					7	14.7 (5.0)
Acute tryptophan depletion	7	10.5 (1.6)	7	0.9 (0.3)					7	0.9 (0.3)

https://doi.org/10.1192/bjp.bp.110.084178 Published online by Cambridge University Press

balanced amino acid mixture produced a significant decrease in [^{11}C]raclopride BP_{ND} values (440 voxels, peak $t=6.2$), consistent with an effect of intranasal cocaine on extracellular dopamine levels with peak effects in the ventral striatum and post-commissural putamen (Fig. 1(a) and online Table DS1(a)). Compared with this effect of cocaine alone (nutritionally balanced amino acid mixture + cocaine), cocaine in the low 5-HT session (acute tryptophan depletion + cocaine) yielded significantly greater changes in [^{11}C]raclopride BP_{ND} values, particularly in dorsal aspects of the anterior and posterior putamen, both left and right, as well as bilateral caudate (208 voxels, peak $t=5.4$) (Fig. 1(b) and online Table DS1(b)). A comparison of the effect of cocaine plus acute tryptophan depletion to the placebo test session (nutritionally balanced amino acid mixture + placebo) yielded decreased [^{11}C]raclopride BP_{ND} values throughout the entire striatum (2867 voxels, peak $t=9.2$) (Fig. 1(c) and online Table DS1(c)).

Experiment 2

Acute tryptophan depletion alone did not decrease [^{11}C]raclopride BP_{ND} values anywhere. In comparison, the voxel-wise analysis indicated that acute tryptophan depletion alone, as compared with the nutritionally balanced amino acid mixture, increased [^{11}C]raclopride BP_{ND} in ventrolateral aspects of the right posterior putamen and bilateral anterior putamen (141 voxels, peak $t=5.4$) (Fig. 2 and online Table DS1(d)).

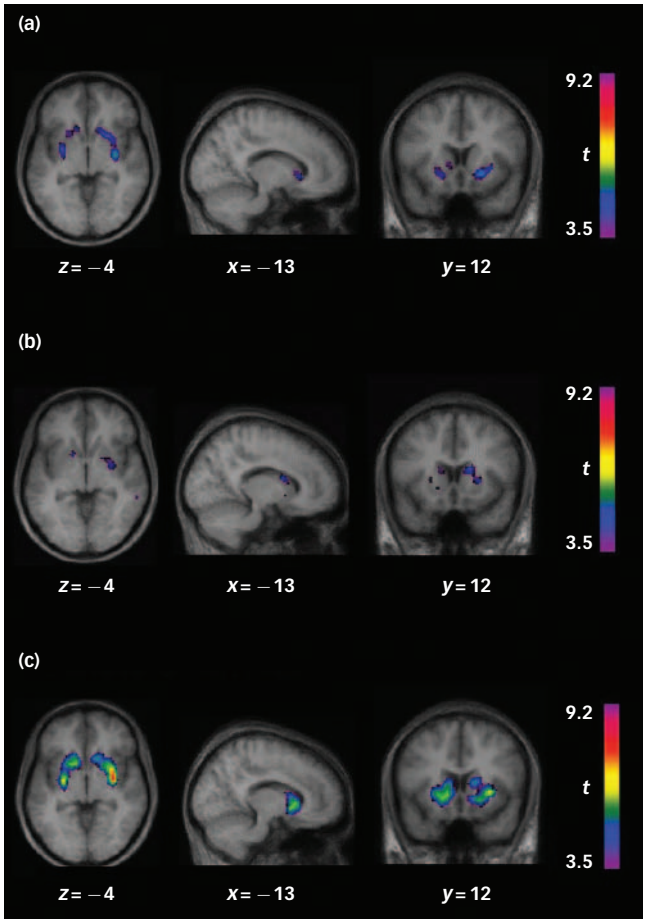

Fig. 1 Three *t*-maps.

(a) Decreased [^{11}C]raclopride BP_{ND} in nutritionally balanced amino acid mixture + cocaine compared with nutritionally balanced amino acid mixture + placebo. (b) Decreased [^{11}C]raclopride BP_{ND} in acute tryptophan depletion + cocaine compared with nutritionally balanced amino acid mixture + cocaine. (c) Decreased [^{11}C]raclopride BP_{ND} in acute tryptophan depletion + cocaine compared with nutritionally balanced amino acid mixture + placebo.

Behavioural data

Cocaine produced its prototypical subjective effects, as reflected by time × condition interactions for the total craving score ($F(3.2, 28.8)=3.66$, $P\leqslant0.022$) as well as drug wanting ($F(3.79, 34.1)=4.68$, $P\leqslant0.005$), drug desire ($F(3.53, 31.8)=3.35$, $P\leqslant0.025$), liking ($F(2.60, 23.4)=5.38$, $P\leqslant0.008$), high ($F(2.53, 22.8)=5.59$, $P\leqslant0.007$), euphoria ($F(2.47, 22.2)=4.14$, $P\leqslant0.023$) and rush ($F(2.97, 26.7)=3.58$, $P\leqslant0.027$) (Table 3). On the low 5-HT test session the cocaine-induced craving response was augmented as reflected by greater increases in cocaine-induced drug wanting ($t(9)=-2.67$, $P\leqslant0.025$, Fig. 3) and the total craving score ($t(9)=2.28$, $P\leqslant0.049$, Table 3). Cocaine's effects on drug liking, high, euphoria and rush, in comparison, were unaltered following acute tryptophan depletion ($P\geqslant0.05$, Table 3). In Experiment 2, acute tryptophan depletion alone did not produce any significant changes in subjective measures as indicated by the absence of significant time × condition interactions on the VAS scales ($F(4, 12)\leqslant2.1$, $P\geqslant0.14$).

Discussion

Main findings

The present findings provide the first demonstration in humans that lowered 5-HT transmission increases cocaine-induced craving states and dopamine responses. These observations may suggest a mechanism by which individuals with disorders associated with

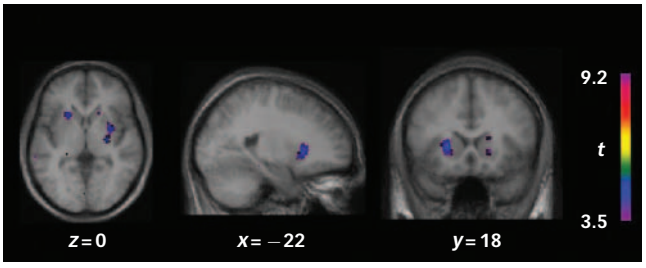

Fig. 2 A *t*-map illustrating increased [^{11}C]raclopride BP_{ND} in acute tryptophan depletion compared with nutritionally balanced amino acid mixture, indicating decreases in dopamine release.

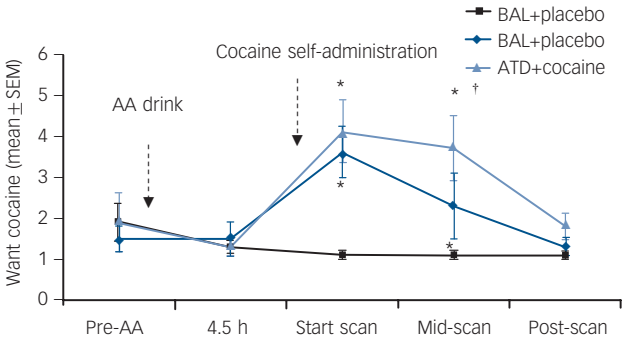

Fig. 3 Cocaine-increased self-reports of 'want cocaine'.

*Significantly different from nutritionally balanced amino acid (AA) mixture + placebo; †significantly different from nutritionally balanced amino acid mixture + cocaine. ATD, acute tryptophan depletion; BAL, nutritionally balanced amino acid mixture.

https://doi.org/10.1192/bjp.bp.110.084178 Published online by Cambridge University Press

Table 3 Subjective effects in Experiment 1

	Subjective effects, mean (s.e.m.)				
	Pre-amino acid	4.5 h	Start scan	Mid-scan	Post-scan
High**					
Nutritionally balanced amino acid mixture + placebo	1.4 (0.2)	1.2 (0.1)	1.4 (0.2)	1.1 (0.1)	1.1 (0.1)
Nutritionally balanced amino acid mixture + cocaine	1.8 (0.6)	1.1 (0.1)	3.8 (0.6)††	2.3 (0.4)†	1.4 (0.2)
Acute tryptophan depletion + cocaine	1.4 (0.2)	1.1 (0.1)	4.7 (0.9)††	3.1 (0.5)††	1.3 (0.2)
Like drug**					
Nutritionally balanced amino acid mixture + placebo	1.9 (0.5)	1.4 (0.2)	1.4 (0.2)	1.2 (0.1)	1.1 (0.1)
Nutritionally balanced amino acid mixture + cocaine	1.4 (0.2)	1 (0)†	4.1 (0.5)††	2.5 (0.4)	1.8 (0.3)
Acute tryptophan depletion + cocaine	2 (0.6)	1.6 (0.4)	5.2 (1.0)††	3.7 (0.6)††	2 (0.4)†
Rush*					
Nutritionally balanced amino acid mixture + placebo	2 (0.4)	2 (0.5)	1.5 (0.2)	1.2 (0.1)	1.3 (0.2)
Nutritionally balanced amino acid mixture + cocaine	2.2 (0.5)	2.1 (0.8)	4 (0.4)†††	2.1 (0.3)†	1.9 (0.4)
Acute tryptophan depletion + cocaine	1.8 (0.3)	1.6 (0.2)	4.4 (0.9)†	3.1 (0.5)††	1.8 (0.3)
Euphoria*					
Nutritionally balanced amino acid mixture + placebo	1.4 (0.2)	1.1 (0.1)	1.1 (0.1)	1 (0)	1.1 (0.1)
Nutritionally balanced amino acid mixture + cocaine	1.5 (0.2)	1.1 (0.1)	2.4 (0.4)†	1.5 (0.2)†	1.2 (0.1)
Acute tryptophan depletion + cocaine	1.7 (0.2)	1.2 (0.1)	3.6 (0.8)	2.1 (0.4)††	1.2 (0.1)
Want cocaine**					
Nutritionally balanced amino acid mixture + placebo	1.9 (0.5)	1.3 (0.1)	1.1 (0.1)	1.1 (0.1)	1.1 (0.1)
Nutritionally balanced amino acid mixture + cocaine	1.5 (0.3)	1.5 (0.4)	3.6 (0.6)††	2.3 (0.8)	1.3 (0.2)
Acute tryptophan depletion + cocaine	1.9 (0.7)	1.3 (0.2)	4.1 (0.8)††	3.7 (0.8)††‡	1.8 (0.3)
Total craving score*					
Nutritionally balanced amino acid mixture + placebo	1.3 (0.1)	1.1 (0.1)	1.1 (0.1)	1.1 (0.1)	1.1 (0.1)
Nutritionally balanced amino acid mixture + cocaine	1.2 (0.1)	1.3 (0.2)	2.8 (0.5)††	2.2 (0.8)	1.2 (0.1)
Acute tryptophan depletion + cocaine	1.5 (0.4)	1.2 (0.1)	3.3 (0.6)††	2.9 (0.7)†‡	1.5 (0.2)

*Treatment × time interaction, significant at $P \leqslant 0.05$, **$P \leqslant 0.01$; †significantly different from nutritionally balanced amino acid mixture + placebo, at $P=0.05$, $P \leqslant 0.01$, $P \leqslant 0.001$; ‡significantly different from nutritionally balanced amino acid mixture + cocaine, significant at $P \leqslant 0.05$.

low 5-HT[1–3] are at increased risk for drug-seeking behaviour and the progression to substance misuse.

The present findings are supported by considerable preclinical evidence for 5-HT/dopamine interactions in the nigrostriatal and mesolimbic dopamine systems. Serotonin neurons in the raphe nuclei project to dopamine cell bodies and terminal regions including the ventral tegmental area, substantia nigra and ventral plus dorsal striatum.[26,27] Inhibitory effects of 5-HT on both the behavioural and dopaminergic effects of cocaine have been reported. For example, administration of a tryptophan-deficient diet to rats significantly increases locomotor activity, conditioned place preference and dopamine responses to amphetamine and cocaine.[7,28] In comparison, increasing 5-HT neurotransmission has the opposite effect, diminishing cocaine-induced striatal dopamine responses and self-administration behaviour.[8,9]

In the present study, acute tryptophan depletion alone did not increase dopamine release, consistent with the hypothesis that the lowered 5-HT was augmenting the cocaine's effects. In comparison, acute tryptophan depletion alone did elicit evidence of decreased dopamine release, particularly within the ventrolateral putamen. In animal models, the predominant effect of 5-HT is to inhibit dopamine cell firing and release, although this occurs most consistently within the substantia nigra and dorsal striatum.[4,29] In comparison, some evidence suggests that 5-HT can increase resting dopamine transmission in more ventral regions. For example electrical stimulation of the dorsal raphe increases the cell firing of most dopamine neurons in the ventral tegmental area,[29] whereas 5-HT depletion leads to decreased ventral tegmental area dopamine cell firing.[6]

The dopaminergic effects of cocaine and acute tryptophan depletion demonstrated some regional specificity, and the relevant neuroanatomy has been studied in depth.[30] The ventral striatum receives dense input from the amygdala, hippocampus and limbic cortex, and dopamine transmission within this area is thought to influence the salience of, and sustained interest in, motivationally relevant stimuli such as rewards.[31–36] The dorsal striatum, in comparison, receives less input from limbic regions and more from the associative and sensorimotor cortical areas. Dopamine transmission within the dorsal striatum has been implicated in the regulation of stimulus–response habit learning.[37,38] In primates, these subdivisions are not sharply delineated, raising the possibility that limbic system-mediated incentive motivational processes extend in a gradation from ventromedial striatum through to more dorsal aspects.[30] If low 5-HT states augment drug-induced dopamine responses within the dorsal striatum, the result might be enhanced motivation to obtain drug reward and susceptibility to compulsive, habit-like drug-seeking behaviour.

Limitations

The conclusions suggested by the present study should be interpreted in light of the following considerations. First, the challenge dose of cocaine was modest, 1.0 mg/kg taken intranasally. However, using this dose protected against ceiling effects, increasing our ability to identify augmentations of the drug-induced dopamine response as a result of acute tryptophan depletion. Second, the sample sizes are modest, but within the commonly accepted range for assessing pharmacological challenges within participants. Third, cocaine and 5-HT can be vasoconstrictors. Altered cerebral blood flow following acute tryptophan depletion or cocaine could produce confounding effects on the observed changes in BP_{ND}. However, the effects of cocaine plus acute tryptophan depletion were synergistic rather than antagonistic. Moreover, simulation studies indicate that even large changes in blood flow have negligible effects on receptor ligand binding, as measured by the method used here.[24] Fourth, in a previous study conducted in individuals who were cocaine dependent, acute tryptophan depletion was reported to leave cocaine-induced craving unaltered and to decrease the drug's

https://doi.org/10.1192/bjp.bp.110.084178 Published online by Cambridge University Press

euphorigenic effects.[39] The apparent discrepancy with the current results may be explained by differences in participant population (dependent *v.* non-dependent) or dose of cocaine administered (2.0 *v.* 1.0 mg/kg). For example, as the authors noted, the diminished high may have been secondary to an acute tryptophan depletion-induced increase in cocaine-elicited anxiety. In the present study with a lower dose of cocaine, significant anxiogenic responses were not produced. The ability of other serotonergic manipulations to influence cocaine's behavioural effects has also been dependent on the cocaine dose, both in humans and in laboratory animals.[11,12] Finally, 15 of the 17 participants were male, precluding our ability to test for potential gender differences.

Implications

In conclusion, the present study's primary findings were that, in humans, a low 5-HT state diminished resting limbic dopamine levels and augmented cocaine-induced striatal dopamine responses plus the desire to use the drug. Although this is the first study in humans of the effects of lowered 5-HT transmission on dopamine release, and the results will require replication, the observed combination of effects might delineate monoamine interactions within human brain and identify a mechanism by which individuals with low serotonergic tone are at elevated risk for substance misuse and various comorbid disorders.

Sylvia M. L. Cox, PhD, **Chawki Benkelfat**, MD, DERBH, Department of Psychiatry and Department of Neurology and Neurosurgery; **Alain Dagher**, MD, Department of Neurology and Neurosurgery; **J. Scott Delaney**, MD, Department of Emergency Medicine; **France Durand**, MSc, **Theodore Kolivakis**, MD, **Kevin F. Casey**, MSc, Department of Psychiatry; **Marco Leyton**, PhD, Department of Psychiatry and Department of Neurology and Neurosurgery, McGill University, Montréal, Québec, Canada

Correspondence: Marco Leyton, PhD, Department of Psychiatry, McGill University, 1033 Pine Avenue West, Montreal, Quebec, Canada, H3A 1A1. Email: marco.leyton@mcgill.ca

First received 5 Jul 2010, final revision 5 Nov 2010, accepted 9 Dec 2010

Funding

This work was supported by an operating grant from the Canadian Institutes of Health Research to M.L. (MOP–36429). M.L. and C.B. are both recipients of research chairs from McGill University. S.M.L.C. received a fellowship from the McGill University Health Centre.

Acknowledgements

We thank Franceen Lenoff, Kathleen Auclair, Sam McKenzie and the PET technicians at the McConnell Brain Imaging Centre for excellent technical assistance, and the Emergency Medicine Services physicians for their valuable medical support.

References

1 Virkkunen M, Rawlings R, Tokola R, Poland RE, Guidotti A, Nemeroff C, et al. CSF biochemistries, glucose metabolism, and diurnal activity rhythms in alcoholic, violent offenders, fire setters, and healthy volunteers. *Arch Gen Psychiatry* 1994; **51**: 20–7.

2 Leyton M, Okazawa H, Diksic M, Paris J, Rosa P, Mzengeza S, et al. Brain regional alpha-[11C]methyl-L-tryptophan trapping in impulsive subjects with borderline personality disorder. *Am J Psychiatry* 2001; **158**: 775–82.

3 Seo D, Patrick CJ, Kennealy PJ. Role of serotonin and dopamine system interactions in the neurobiology of impulsive aggression and its comorbidity with other clinical disorders. *Aggress Violent Behav* 2008; **13**: 383–95.

4 Kapur S, Remington G. Serotonin-dopamine interaction and its relevance to schizophrenia. *Am J Psychiatry* 1996; **153**: 466–76.

5 Guan XM, McBride WJ. Serotonin microinfusion into the ventral tegmental area increases accumbens dopamine release. *Brain Res Bull* 1989; **23**: 541–7.

6 Minabe Y, Emori K, Ashby CR Jr. The depletion of brain serotonin levels by para-chlorophenylalanine administration significantly alters the activity of midbrain dopamine cells in rats: an extracellular single cell recording study. *Synapse* 1996; **22**: 46–53.

7 Carta M, Collu M, Fadda F, Stancampiano R. Augmented cocaine-induced accumbal dopamine efflux, motor activity and place preference in rats fed with a tryptophan-deficient diet. *Neurosci Lett* 2006; **401**: 125–9.

8 Czoty PW, Ginsburg BC, Howell LL. Serotonergic attenuation of the reinforcing and neurochemical effects of cocaine in squirrel monkeys. *J Pharmacol Exp Ther* 2002; **300**: 831–7.

9 Molina PE, Ahmed N, Gatley J, Volkow ND, Abumrad NN. L-tryptophan attenuation of the dopaminergic and behavioral responses to cocaine. *Life Sci* 2001; **69**: 1897–906.

10 Fletcher PJ, Korth KM, Chambers JW. Selective destruction of brain serotonin neurons by 5,7-dihydroxytryptamine increases responding for a conditioned reward. *Psychopharmacology (Berl)* 1999; **147**: 291–9.

11 Walsh SL, Cunningham KA. Serotonergic mechanisms involved in the discriminative stimulus, reinforcing and subjective effects of cocaine. *Psychopharmacology (Berl)* 1997; **130**: 41–58.

12 Peltier R, Schenk S. Effects of serotonergic manipulations on cocaine self-administration in rats. *Psychopharmacology (Berl)* 1993; **110**: 390–4.

13 Cox SM, Benkelfat C, Dagher A, Delaney JS, Durand F, McKenzie SA, et al. Striatal dopamine responses to intranasal cocaine self-administration in humans. *Biol Psychiatry* 2009; **65**: 846–50.

14 First MB, Spitzer RL, Gibbon M, Williams JBW. *Structural Clinical Interview for DSM-IV-TR Axis I Disorders: Research Version, Non-patient Edition. (SCID-I/NP)*. Biometrics Research, 2002.

15 Heatherton TF, Kozlowski LT, Frecker RC, Fagerstrom KO. The Fagerstrom Test for Nicotine Dependence: a revision of the Fagerstrom Tolerance Questionnaire. *Br J Addict* 1991; **86**: 1119–27.

16 Young SN, Smith SE, Pihl RO, Ervin FR. Tryptophan depletion causes a rapid lowering of mood in normal males. *Psychopharmacology (Berl)* 1985; **87**: 173–7.

17 Sled JG, Zijdenbos AP, Evans AC. A nonparametric method for automatic correction of intensity nonuniformity in MRI data. *IEEE Trans Med Imaging* 1998; **17**: 87–97.

18 Talairach J, Tournoux P. *Co-planar Stereotaxic Atlas of the Human Brain*. Georg Thieme Verlag, 1988.

19 Collins DL, Neelin P, Peters TM, Evans AC. Automatic 3D intersubject registration of MR volumetric data in standardized Talairach space. *J Comput Assist Tomogr* 1994; **18**: 192–205.

20 Evans AC, Marrett S, Neelin P, Collins L, Worsley K, Dai W, et al. Anatomical mapping of functional activation in stereotactic coordinate space. *Neuroimage* 1992; **1**: 43–53.

21 Costes N, Dagher A, Larcher K, Evans AC, Collins DL, Reilhac A. Motion correction of multi-frame PET data in neuroreceptor mapping: simulation based validation. *Neuroimage* 2009; **47**: 1496–505.

22 Gunn RN, Lammertsma AA, Hume SP, Cunningham VJ. Parametric imaging of ligand-receptor binding in PET using a simplified reference region model. *Neuroimage* 1997; **6**: 279–87.

23 Laruelle M. Imaging synaptic neurotransmission with in vivo binding competition techniques: a critical review. *J Cereb Blood Flow Metab* 2000; **20**: 423–51.

24 Aston JA, Gunn RN, Worsley KJ, Ma Y, Evans AC, Dagher A. A statistical method for the analysis of positron emission tomography neuroreceptor ligand data. *Neuroimage* 2000; **12**: 245–56.

25 Worsley KJ, Marrett S, Neelin P, Vandal AC, Friston KJ, Evans AC. A unified statistical approach for determining significant signals in images of cerebral activation. *Hum Brain Mapp* 1996; **4**: 58–73.

26 Hervé D, Pickel VM, Joh TH, Beaudet A. Serotonin axon terminals in the ventral tegmental area of the rat: fine structure and synaptic input to dopaminergic neurons. *Brain Res* 1987; **435**: 71–83.

27 Fibiger HC, Miller JJ. An anatomical and electrophysiological investigation of the serotonergic projection from the dorsal raphe nucleus to the substantia nigra in the rat. *Neuroscience* 1977; **2**: 975–87.

28 Carta M, Fadda F, Stancampiano R. Tryptophan-deficient diet increases the neurochemical and behavioral response to amphetamine. *Brain Res* 2006; **1094**: 86–91.

29 Gervais J, Rouillard C. Dorsal raphe stimulation differentially modulates dopaminergic neurons in the ventral tegmental area and substantia nigra. *Synapse* 2000; **35**: 281–91.

30 Haber SN, Kim KS, Mailly P, Calzavara R. Reward-related cortical inputs define a large striatal region in primates that interface with associative cortical connections, providing a substrate for incentive-based learning. *J Neurosci* 2006; **26**: 8368–76.

https://doi.org/10.1192/bjp.bp.110.084178 Published online by Cambridge University Press

31 Robinson, TE, Berridge KC. Review. The incentive sensitization theory of addiction: some current issues. *Philos Trans R Soc Lond B Biol Sci* 2008; **363**: 3137–46.

32 Grace AA, Floresco SB, Goto Y, Lodge DJ. Regulation of firing of dopaminergic neurons and control of goal-directed behaviors. *Trends Neurosci* 2007; **30**: 220–7.

33 Koob GF, Volkow ND. Neurocircuitry of addiction. *Neuropsychopharmacology* 2010; **35**: 217–38.

34 Leyton, M. The neurobiology of desire: dopamine and the regulation of mood and motivational states in humans. In *Pleasures of the Brain* (eds ML Kringelbach, KC Berridge). Oxford University Press, 2009.

35 Schultz W. Behavioral dopamine signals. *Trends Neurosci* 2007; **30**: 203–10.

36 Phillips AG, Vacca G, Ahn S. A top-down perspective on dopamine, motivation and memory. *Pharmacol Biochem Behav* 2008; **90**: 236–49.

37 McDonald RJ, White NM. A triple dissociation of memory systems: hippocampus, amygdala, and dorsal striatum. *Behav Neurosci* 1993; **107**: 3–22.

38 Everitt BJ, Robbins TW. Neural systems of reinforcement for drug addiction: from actions to habits to compulsion. *Nat Neurosci* 2005; **8**: 1481–9.

39 Aronson SC, Black JE, McDougle CJ, Scanley BE, Jatlow P, Kosten TR, et al. Serotonergic mechanisms of cocaine effects in humans. *Psychopharmacology (Berl)* 1995; **119**: 179–85.

EXTRA CONTENT ONLINE

extra

Buried alive!

Cyrus Abbasian

Sadeq Hedayat (1903–1951), a 20th-century pioneer of modern Persian literature, was particularly notorious for his Kafkaesque psycho-fictions. Franz Kafka was undoubtedly a great influence on Hedayat, who was the first Persian translator of *The Metamorphosis* (1915).

Hedayat's tortured soul informed his detailed writings on suicidal tendencies, especially in the short story *Buried Alive* (1930). He alludes to the comforting notion of an imminent death: 'No one decides to commit suicide . . . suicide is within some of us, as an essence and disposition'. He adds, 'it's terrifying when death doesn't want and rejects us.' Despite the social stigma attached to suicide within the Persian culture, Hedayat was uplifted and comforted by the option of an imminent death. During a walk in a cemetery he writes, 'I envied the dead . . . I consider death as joyous; a miracle that no one is given easily.'

Hedayat first attempted suicide by jumping into the Seine before writing *Buried Alive*. Therein he further details the processes preceding suicide, outlining the desire of inducing grave physical illness before overdosing on drugs. He fantasises about slowly dying after taking aspirin with opium, preoccupied with the need to 'sense death properly' and the reaction of those who discover his corpse.

Finally, in 1951, a series of adverse events, including financial and emotional ruin, contribute to his ending. He meticulously planned and executed his suicide and was buried at Père Lachaise Cemetery in Paris. Although no final note was left, Hedayat's writings remain his testament.

Cyrus Abbasian is a consultant psychiatrist. The translations are the author's own from the original Persian.

The British Journal of Psychiatry (2011) 199, 397. doi: 10.1192/bjp.bp.111.095844

https://doi.org/10.1192/bjp.bp.110.084178 Published online by Cambridge University Press