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Archival Report

Striatal Dopamine Release in Response to Morphine: A [^{11}C]Raclopride Positron Emission Tomography Study in Healthy Men

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ABSTRACT

BACKGROUND: Preclinical and human positron emission tomography studies have produced inconsistent results regarding the effects of opioids on mesolimbic dopamine (DA). Here, we quantify striatal DA release (measured by [^{11}C]raclopride displacement) in response to an intravenous infusion of morphine, and its relationship with morphine-induced subjective effects, in healthy, nondependent opioid–experienced participants.

METHODS: Fifteen healthy male participants were initially included. Sessions were on separate days. On session 1, participants received intravenous morphine (10 mg/70 kg) in the clinic to ensure tolerability. Participants without adverse reactions (n = 10) then received intravenous morphine and placebo (saline) sessions, in counterbalanced order, while undergoing [^{11}C]raclopride positron emission tomography scans. Subjective and physiological responses were assessed. Region-of-interest and voxelwise image analyses were used to assess changes in [^{11}C]raclopride nondisplaceable binding potential.

RESULTS: Morphine produced marked subjective and physiological effects and induced a significant decrease in [^{11}C]raclopride nondisplaceable binding potential, particularly in the nucleus accumbens and globus pallidus, where the change in [^{11}C]raclopride nondisplaceable binding potential was approximately 9%. However, the subjective effects of morphine did not show a simple pattern of correlation with DA release.

CONCLUSIONS: This is, to our knowledge, the first study providing in vivo human evidence that DA transmission in the ventral striatum is affected by morphine. Further studies are required to fully delineate the DA contribution to the reinforcing effects of opioids.

Keywords: [^{11}C]Raclopride, Dopamine, Morphine, Opioids, PET, Ventral striatum

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Opioids have been used by humans for their analgesic and euphorigenic effects for thousands of years. In the last century, natural and synthetic opioids, including morphine, oxycodone, and fentanyl, have become mainstays of treatment for acute and chronic pain. The drastic increase in rates of prescription opioid use, particularly since the 1990s, posed the challenge of balancing the analgesic properties of opioids against their well-documented abuse potential resulting from their potent rewarding properties in vulnerable individuals.

Currently, the nonmedical use of prescription as well as illicit opioids represents a major public health crisis in the United States (1). Because the potent reinforcing properties of opioid agonists are combined with respiratory depressant effects, the increased prevalence of nonmedical opioid use has resulted in dramatically increased mortality, reaching 60,000 deaths annually (2). The opioid epidemic requires broad implementation of available treatments for opioid use disorder (OUD), but also prompts a need for mechanistically novel interventions for pain.

Major advances have been made in the neuroscience of opioid actions and OUD, but the mechanisms underlying the effects of morphine and other opioids, particularly with regard to their reinforcing properties, are still highly debated (3–5). Preclinical studies indicate that opioids act in the ventral tegmental area via Gi/o-coupled mu opioid receptors to disinhibit dopaminergic neurons by attenuating inhibitory gamma-aminobutyric acidergic tone. This promotes burst firing of dopaminergic neurons and enhances dopamine (DA) release in their terminal fields (6,7). In vivo microdialysis data also demonstrate that opioid administration increases DA release in the nucleus accumbens (NAc) (8).

While this dopaminergic response has been evidenced in animal investigations, modulation of mesolimbic DA transmission by opioids remains controversial in humans. Two previous [^{11}C]raclopride positron emission tomography (PET) studies have evaluated changes in extracellular DA release following the administration of opioids. These studies failed to

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detect increased striatal DA release in response to opioids, an effect reported for other addictive agents [for a review, see Van Ree and Ramsey (9) and Gerrits *et al.* (10)]. Specifically, an intravenous dose of 50-mg heroin did not result in a measurable effect on striatal DA levels in heroin-dependent patients, despite producing marked euphoria (11). Similarly, heroin administration, as well as the expectation of a heroin reward, was not associated with DA release in patients with OUD receiving methadone or buprenorphine (12).

However, assessing the magnitude of DA release in individuals with a prolonged exposure to opioids may be misleading, given that chronic drug exposure induces long-term adaptations within the dopaminergic system. These neuroadaptive processes result in a hypodopaminergic state, indexed experimentally by a blunted dopaminergic response. This phenomenon has been observed in opioid-dependent individuals (13), as well as in individuals with other substance use disorders (14–17), and is also supported by extensive preclinical data [for a review, see Fields and Margolis (18)]. Moreover, studies in rodents suggest that the differential dopaminergic response observed in opioid-naïve versus opioid-dependent animals may underlie a different contribution of DA to opioid reinforcement, depending on opioid exposure state (19,20). To date, the role of the dopaminergic response in opioid reinforcement remains the subject of debate [(9,10,21–27), but see also (28–31)]. Thus, a better understanding of the dopaminergic response to opioids in the human brain is needed.

To begin addressing this set of questions, we hypothesized that in nondependent opioid–experienced participants, morphine administration would increase striatal DA release compared with placebo, although to a lesser degree than that elicited by substances acting directly on the dopaminergic system, as also indicated by preclinical studies (6). We used PET and the $D_{2/3}$ receptor–preferring radioligand [^{11}C]raclopride to test this hypothesis. Together with measures of DA release, we evaluated the subjective effects induced by intravenous infusion of morphine, with the aim of exploring the relationship between those and DA release.

METHODS AND MATERIALS

Participants

Fifteen healthy male participants who had previously received oral prescription opioid analgesics were recruited from the community via flyers, newspapers, and Internet advertisements (see Supplemental Table S1 for demographics). Participants provided written informed consent and underwent a thorough medical examination prior to enrollment in the study. Further details are provided in Supplement.

Experimental Design

Participants underwent three single-blinded infusion sessions on separate days, separated by approximately 1 week. At the start of each session, a urine drug screen (iScreen One Step 10 Panel Dip Card; Alere Toxicology, Waltham, MA) (testing for amphetamine, phencyclidine, buprenorphine, methadone, morphine, oxycodone, Δ^9-tetrahydrocannabinol, cocaine, methamphetamine, 3,4-methylenedioxy-methamphetamine), a breathalyzer test, and carbon monoxide monitoring were performed to exclude the use of psychoactive drugs of abuse, alcohol, and recent tobacco smoking (carbon monoxide ≥15 ppm disqualified the subject from the study). Cotinine levels were assessed for participants reporting e-cigarette use (cotinine ≥1 ng/mL disqualified the subject from the study). As a safety measure, food and beverages (except for water) were discontinued at least 4 hours prior to imaging studies.

A baseline infusion session (first session) was performed in the clinic to ensure that participants tolerated the morphine infusion without experiencing nausea or marked sedation. During this session, which lasted approximately 5 hours, participants received, in a fixed order, a placebo infusion (normal saline) followed by an intravenous challenge of morphine (10 mg/70 kg over 1 minute; morphine concentration 2 mg/mL). This is at the high end of doses used in clinical practice to achieve analgesia. Pre- and postinjection, the following measures were acquired: 1) subjective responses as measured by the Drug Effects Questionnaire (DEQ) (32), which assessed drug-liking, drug-wanting, high, and feel drug; and 2) physiological response, including respiratory rate, oxygen saturation, and pupil response to light. The DEQ was administered at baseline and at 5, 15, 30, 45, 60, 75, and 90 minutes after each injection. The pupillary constriction test was modified from a previously reported method (33). In brief, participants were instructed to focus on a fixation point and to avoid blinking while the pupilometer (PLR-200 Pupillometer; NeurOptics, Laguna Hills, CA) was positioned over the eye by the operator. The resting pupil diameter of each eye was measured for 2 seconds every 20 seconds, for a total of 1 minute. The test was performed at baseline and at 10, 20, and 30 minutes postinjection. Participants were also monitored throughout the experimental session with pulse oximetry for safety. Following the first session, 5 participants were excluded from the study because they experienced adverse effects (mostly nausea) in response to morphine.

PET and Magnetic Resonance Imaging Scans

In the second and third sessions, which lasted approximately 3.5 hours each, the remaining 10 participants received, in counterbalanced, randomized order, an intravenous infusion of morphine (10 mg/70 kg) or an equivalent volume of normal saline over 20 to 30 seconds, while undergoing a PET scan with [^{11}C]raclopride. Participants were scanned supine with their head held in place using a custom-made thermoplastic facemask fixation system to minimize head movements during scanning. Subjective effects were assessed using the DEQ at baseline and every 10 minutes during the scan, while vital signs and electrocardiograms were collected at baseline and every 5 minutes during the scan. Details on the PET scan protocol are provided in the Supplement.

For structural reference, 3-dimensional T1-weighted magnetic resonance imaging (MRI) scans were obtained prior to the PET sessions using a 3T MRI scanner (GE Healthcare, Milwaukee, WI).

Data Processing

The raw PET data were acquired in 3-dimensional mode, reconstructed into dynamic time frames of variable duration

(0.5–5 minutes), and then co-registered with standard T1-weighted (magnetization prepared rapid acquisition gradient-echo) MRI scans using PMOD version 2.8.5 (PMOD Technologies Ltd., Zürich, Switzerland). All PET images were then resliced using SPM8 (Wellcome Trust Centre for Neuroimaging, London, UK). Parametric binding potential (BP) images were obtained using the Simple Reference Tissue Model 2 (34), and the cerebellum was used as the reference region to derive a quantitative estimate of BP relative to the nondisplaceable compartment. The PET data from the placebo session were used as a measure of baseline raclopride nondisplaceable BP (BP_{ND}). Morphine-induced changes in BP (ΔBP_{ND}) in each region of interest (ROI) were computed relative to baseline as % $\Delta BP_{ND} = 100 \times \Delta BP_{ND}$ (placebo – morphine)/BP_{ND} (placebo). Reduction in raclopride binding is attributed to competition with endogenous DA, and the percent change in BP has been shown to be proportional to the magnitude of DA release (35). Further details on PET data processing are provided in the Supplement.

Voxelwise Analysis

Voxelwise parameter estimates of [^{11}C]raclopride were generated using Simple Reference Tissue Model 2, with the cerebellar cortex as reference region. PET images were first analyzed on a pixel-by-pixel basis with PMOD. Next, BP_{ND} changes from placebo to morphine condition were investigated statistically using a paired *t* test in SPM8. Analysis was performed on voxels with BP_{ND} values >1 across participants (which primarily included the striatum). A stringent statistical significance threshold was set by a familywise error (FWE) *p* value < .05, corrected for multiple comparisons at the cluster level using the FWE correction, a cluster-defining threshold $p < .005$ (uncorrected), and a minimum cluster size of 450 voxels (1-mm isotropic), consistent with current statistical standards (36).

ROI Analysis

As a complementary approach to voxelwise analysis, we also performed an additional ROI-level analysis. ROI-based average values are considered more robust than single-voxel measures, as they are less prone to imaging noise. In addition, our ROI analysis was performed in the subject space with ROI defined for each individual, which is more robust to individual differences in regional morphometry. Specifically, subcortical ROIs were defined using an automated segmentation approach (run_first_all command) implemented in FSL version 5.0 (http://www.fmrib.ox.ac.uk/fsl) (37). Four a priori regions were used: the bilateral caudate; putamen; NAc; and pallidum, which included the dorsal pallidum or globus pallidus (GP) (internal and external [GPe] segments). To remove the effect of voxels with low BP_{ND} for each bilateral ROI, the average of the top 10% voxels with the highest BP_{ND} values was estimated. This approach was also motivated by the fact that ROIs are not functionally homogeneous and that the mean of all voxels within the ROI may not optimally represent activity when activated voxels are grouped with inactive or deactivated voxels (38–42). For example, the average map of top 10% voxels indicated that the voxels with the highest BP_{ND} values in the pallidum ROI were located primarily within the GPe; thus, we refer to this particular subregion (GPe) when referring to the pallidum ROI in the rest of the article (Supplemental Figure S1).

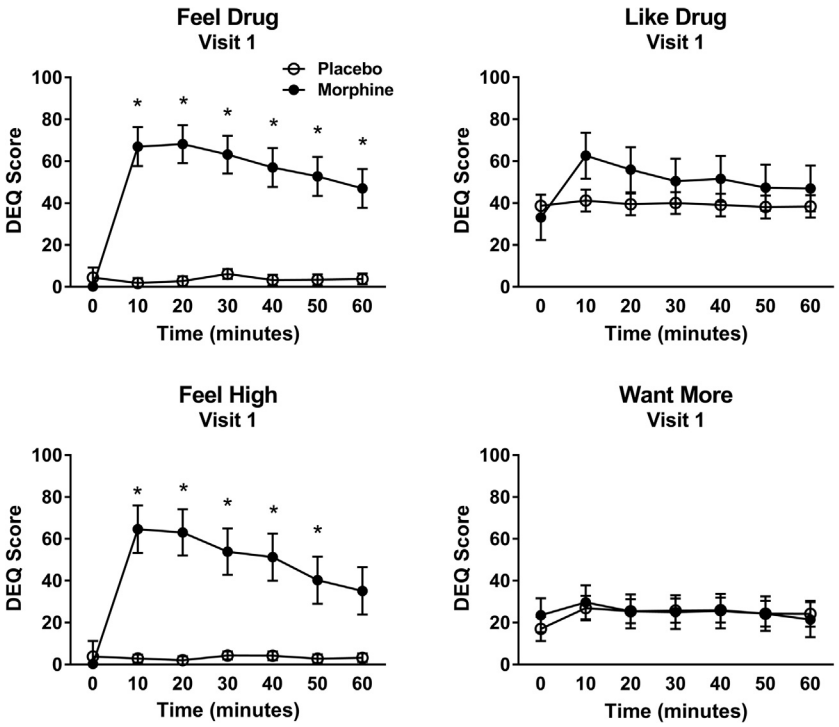

Figure 1. Subjective responses to morphine and placebo outside the scanner environment (visit 1) on the Drug Effects Questionnaire (DEQ). Injection of morphine produced significant condition × time interactions on feel high ($F_{6,56} = 11.52$, $p < .0001$), feel drug ($F_{6,51} = 15.19$, $p < .0001$), and like drug ($F_{6,55} = 2.67$, $p = .02$), with no effect on want more ($F_{6,59} = 0.18$, $p = .98$). Sample size: 10 healthy, nonsmoking men. * indicates the timepoints showing significant differences between morphine and placebo.

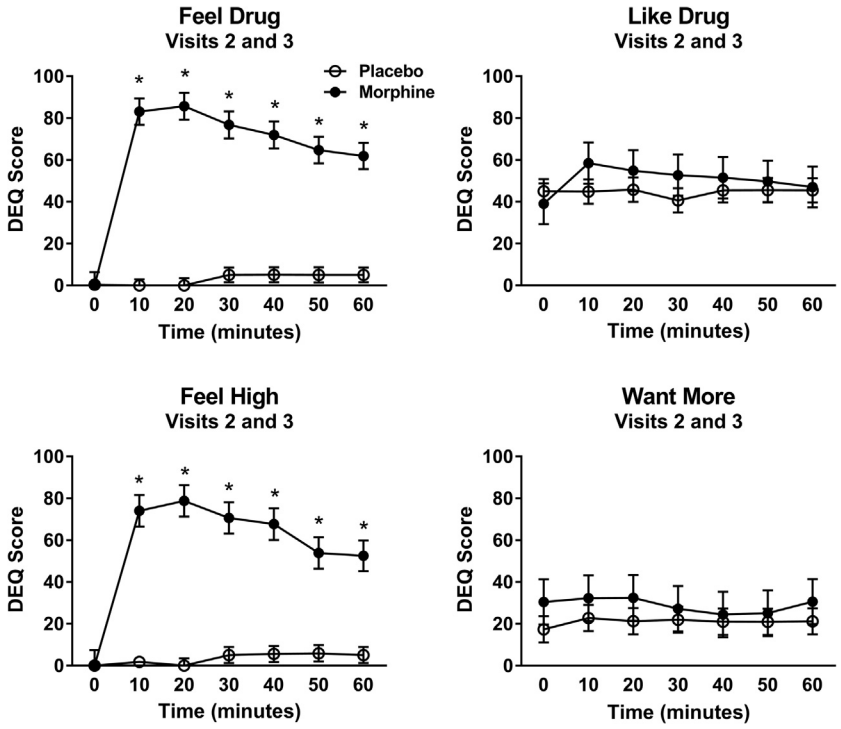

Figure 2. Subjective responses to morphine and placebo inside the scanner environment (visits 2 and 3) on the Drug Effects Questionnaire (DEQ). Injection of morphine produced significant condition × time interactions on feel high ($F_{6,51}$ = 31.19, $p < .0001$) and feel drug ($F_{6,47}$ = 72.32, $p < .0001$), a trend-level condition × time interaction for like drug ($F_{6,61}$ = 2.17, $p = .06$), and no significant effects on want-more ratings ($F_{6,51}$ = 0.51, $p = .80$). Sample size: 10 healthy, nonsmoking men. * indicates the timepoints showing significant differences between morphine and placebo.

Statistical Analysis

All statistical comparisons, apart from voxelwise analyses, were performed using SAS version 9.3 (SAS Institute, Cary, NC).

To determine whether $\%\Delta BP_{ND}$ differed significantly between the placebo and the morphine condition at $p < .05$ (i.e., whether a significant displacement occurred following morphine administration), a one-sample *t* test was performed separately for each ROI. In addition, the absolute BP_{ND} values in each ROI measured were compared between the placebo and morphine conditions using analysis of covariance, with the order of infusion, age, and body mass index as covariates.

Scores on the self-reported DEQ measures were analyzed in two contexts: 1) on the first session (baseline session), in which participants received both placebo and morphine during the same visit outside of the scanner; and 2) between the two following visits, each conducted in the scanner, during which participants received either placebo or morphine during each visit. Data were analyzed only for the 10 subjects who also underwent the PET scan. One included subject had missing DEQ data at several time points during the morphine infusion in the PET scanner. For each context, data were analyzed using a two-way repeated-measures analysis of variance with two within-subject factors: condition (placebo, morphine) and time (seven time points from 0 to 60 minutes). Similarly, the pupillometry data were analyzed with a two-way repeated-measures analysis of variance with two within-subject factors: condition (placebo, morphine) and time (four time points from 0 to 30 minutes), with separate analyses run for the left and right eyes. In all analyses, age and body mass index were included as covariates. All analysis of variance analyses were conducted using the PROC MIXED procedure in SAS.

Correlation analyses between regional changes in [^{11}C]raclopride BP_{ND} and subjective responses were analyzed using the Pearson product-moment correlation coefficient.

RESULTS

Physiological Measures

Injection of morphine produced the expected decrease in pupil diameter, with a significant condition × time interaction for both the right ($F_{3,35}$ = 16.18, $p < .0001$) and left ($F_{3,35}$ = 17.98, $p < .0001$) eyes (Supplemental Figure S2), thus indirectly confirming the acute central effects of morphine.

Subjective Measures

On the first visit, participants showed significant increases in the subjective DEQ measures of feel high (condition × time interaction [$F_{6,56}$ = 11.52, $p < .0001$]), and feel drug (condition × time interaction [$F_{6,51}$ = 15.19, $p < .0001$]), following morphine compared with placebo. The subjective experience of like drug also showed a significant condition × time interaction ($F_{6,55}$ = 2.67, $p = .02$); however, there were no significant differences between morphine and placebo at any of the individual timepoints (all Tukey post hoc tests *p* values > .05). Finally, there was no effect of morphine on the want-more measure (condition × time interaction [$F_{6,59}$ = 0.18, $p = .98$]) (Figure 1). Subjective responses to morphine were similar when participants were in the scanner environment: significant condition × time interactions for feel high ($F_{6,51}$ =

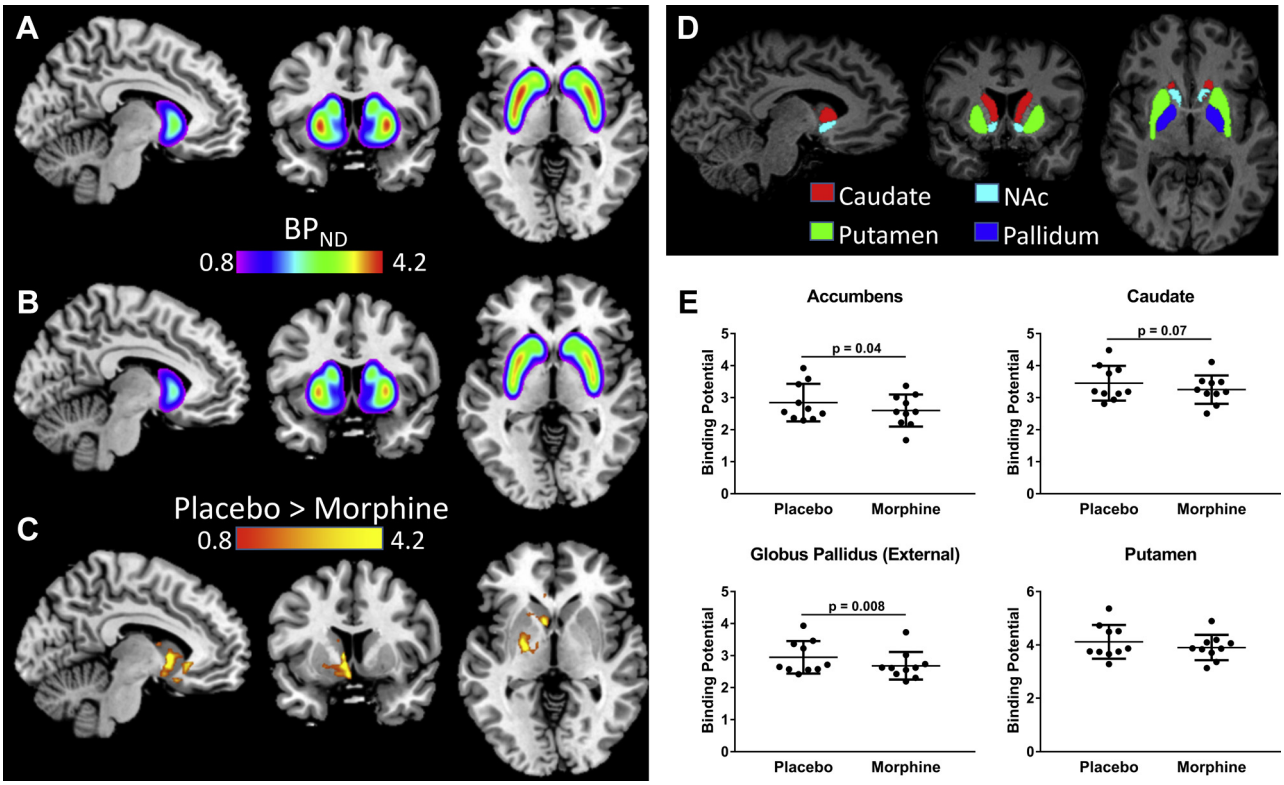

Figure 3. Changes in [^{11}C]raclopride binding potential (BP_{ND}) between the morphine and placebo sessions. **(A, B)** BP_{ND} maps averaged across all participants, coregistered to a common template for the **(A)** morphine and **(B)** placebo infusions. **(C)** Statistical significance (*t* score) map shows a significant decrease in [^{11}C]raclopride BP_{ND} in a large cluster encompassing the putamen, caudate, and external segment of the globus pallidus (GPe) in response to morphine, compared with placebo infusion ($t = 3.5$; p [familywise error] = .005). **(D)** Four a priori regions of interest—caudate, putamen, nucleus accumbens (NAc), and pallidum, which included the dorsal pallidum or globus pallidus (internal and external segments). **(E)** ΔBP_{ND} for [^{11}C]raclopride between placebo and morphine sessions. For the pallidum region of interest, ΔBP_{ND} values refer to the GPe, given that the average map of top 10% voxels indicated that the voxels with the highest BP_{ND} values were located primarily within this subregion of the pallidum. Data are least-square-means (± SEM). Sample size: 10 healthy, nonsmoking, right-handed men. Error bars are SEM.

31.19, $p < .0001$) and feel drug ($F_{6,47} = 72.32$, $p < .0001$) was observed, while no significant effects of condition were found for want-more ratings (condition × time interaction [$F_{6,51} = 0.51$, $p = .80$]). Like-drug measures showed a trend-level condition × time interaction ($F_{6,61} = 2.18$, $p = .06$), but no significant differences between conditions at any of the time points were found (Figure 2).

[^{11}C]Raclopride PET

The voxelwise analysis showed a significant decrease in [^{11}C]raclopride BP_{ND} in a large cluster encompassing the putamen, caudate, and pallidum in response to morphine compared with placebo infusion ($p_{FWE} < .05$) as shown by the averaged statistical maps (Figure 3, Table 1).

The independent ROI analyses corroborated that [^{11}C]raclopride BP_{ND} in the NAc was significantly lower following morphine infusion than placebo infusion ($t_{10} = 2.27$, $p = .04$ [uncorrected]) and in the GPe ($t_{10} = 3.52$, $p = .006$). Differences in [^{11}C]raclopride BP_{ND} in the GPe remained significant ($p = .007$) after partial volume effects correction to account for spillover effects resulting from the limited spatial resolution of the PET camera (see Supplement for partial volume effects correction methodology). There was a trend-level effect in the caudate ($t_{10} = 1.96$; $p = .08$) but there were no significant differences in [^{11}C]raclopride BP_{ND} in the putamen ($t_{10} = 1.6$, $p = .14$) (Figure 3E). An analysis of covariance showed similar results after controlling for age, body mass index, and order of infusion. Specifically, morphine induced a significantly lower [^{11}C]raclopride BP_{ND} compared with placebo in the GPe ($F_{1,9} = 11.5$, $p = .008$; Cohen's $d = 0.67$; 9.0% reduction) and in the NAc ($F_{1,9} = 5.43$; $p = .04$; Cohen's $d = 0.52$; 8.8% reduction). As already observed, a trend-level condition effect was found in the caudate ($F_{1,9} = 4.27$; $p = .07$; Cohen's $d = 0.56$), but no effect of condition was found in the putamen ($F_{1,9} = 3.1$, $p = .11$).

Regional [^{11}C]raclopride BP_{ND} in the morphine and placebo conditions is reported in Table 2.

Correlations Between PET Data and Subjective Responses to Morphine

[^{11}C]Raclopride $\%\Delta BP_{ND}$ was negatively correlated with self-reported drug wanting in the caudate ($r^2 = .51$, $p = .03$) and putamen ($r^2 = .43$, $p = .05$), while only a trend toward significance was observed in the GPe ($r^2 = .39$, $p = .07$) and NAc ($r^2 = .43$, $p = .09$) (Supplemental Figure S3A). $\%\Delta BP_{ND}$ within the GPe was also negatively correlated with subjective ratings

Table 1. Statistics and Spatial Coordinates of Clusters That Demonstrated Significant Differences in [^{11}C]Raclopride BP_{ND} Between the Morphine and Placebo Conditions

	MNI Coordinates, mm			Cluster	PL > MOR
Brain Region	x	y	z	k	*t*
Caudate (Left)[a]	−7	9	3	1489	7.0

The locations of the clusters are based on the coordinates from the MNI stereotactic space (x, y, z). The values correspond to the *t* scores and significance was set at *p* (familywise error) < .005, k ≥ 450.

BP_{ND}, nondisplaceable binding potential; MNI, Montreal Neurological Institute; PL, placebo; MOR, morphine.

[a]The caudate cluster encompasses the caudate body, caudate head, putamen, and pallidum.

of high (r^2 = .62, p = .01) and marginally negatively correlated with self-reported feel-drug measures (r^2 = .44, p = .05) (Supplemental Figure S3B, C). Finally, we did not observe any correlation between drug-liking ratings and [^{11}C]raclopride displacement in any region of interest (data not shown).

DISCUSSION

To our knowledge, this is the first study investigating morphine-induced DA release in mesolimbic areas in nondependent opioid–experienced healthy individuals. Using both ROI and voxelwise analyses, we found that morphine produced a decrease in striatal [^{11}C]raclopride BP, an observation typically interpreted as reflecting an increased release of endogenous DA.

We detected a significant decrease in [^{11}C]raclopride BP_{ND} in response to morphine in the GPe (−9.0% in BP_{ND}) and NAc (−8.8% in BP_{ND}), i.e., in the ventral striatum, which is thought to be most directly linked to reinforcing properties of drugs. Dorsal striatal regions only showed a trend for morphine-induced DA-release. The changes in BP_{ND} in the ventral striatum are of somewhat lower magnitude than commonly reported in similar studies investigating psychostimulants effects in healthy participants (43–45). A contributing factor to the more robust effect of stimulants could be their direct action on the DA transporter (46), but comparison between drug classes is challenging and would require an assessment of the dose-response relationship for the respective class. Increases in DA release similar in magnitude to what we detected, as measured by reduced [^{11}C]raclopride binding, have been reported following acute exposure to alcohol in social drinkers (47–49). Similar findings have also been reported in response to nicotine in smokers (50–52), although these have been less consistent across studies and seem to also be influenced by the sensitivity of the radiotracer used and genetic variation within the DA system (53).

To interpret the magnitude of the BP_{ND} change observed in our study, it is of interest to compare our findings with data obtained from preclinical studies. In their seminal study, Di Chiara and Imperato (6) used in vivo microdialysis to demonstrate that opioids induce DA release but do so to a lesser extent than psychostimulants (<300% vs. 400%–1000%). They also observed that DA release was particularly pronounced in the shell of the NAc, at the terminal site of opioid-sensitive neurons in the ventral tegmental area. Subsequently, in a key bridging study in nonhuman primates, Breier *et al.* (54) found that a fivefold increase in extracellular DA in the striatum was required to produce a 10% decrease in [^{11}C]raclopride binding. The magnitude of BP_{ND} change observed in our study, therefore, seems consistent with the animal literature.

Two previous [^{11}C]raclopride PET studies in opioid-dependent individuals failed to detect increased striatal DA release following acute opioid administration (9,10). Direct comparison with our study is difficult, as the protocols and sample characteristics were substantially different. Sensitivity estimates of [^{11}C]raclopride to changes in extracellular DA concentration are variable, ranging from 8:1 to 44:1 (ratio of % increase in DA and % decrease in [^{11}C]raclopride) (9). Furthermore, the magnitude of DA release is affected by several factors, including the nature of the challenge (54–56) and the drug exposure state (12,13,57,58).

The negative studies enrolled opioid-dependent patients in maintenance therapy with methadone [15 mg/day (9) or 30 mg/day (10)] or buprenorphine [8 mg/day (10)]. These medications were discontinued 24 hours prior to the PET scan, but their long half-life and prolonged repeated dosing could have blunted DA release after the opioid challenge. In addition, patients in the previous, negative studies had a history of opioid dependence. Chronic exposure to opioids may lead to profound changes within the dopaminergic system. Several PET studies in opioid-dependent individuals have shown that striatal $D_{2/3}$ receptor BP is reduced compared with controls [(11,58,59); but see also Gerrits *et al.* (10)]. Studies in rodents have also shown that opioid exposure is associated with a decrease in $D_{2/3}$ receptor binding in the striatum (11,60,61). A similar phenomenon has also been observed in cocaine and methamphetamine users during both early and protracted withdrawal (62–66), as well as in detoxified alcoholic patients (14).

Taken together, our data suggest that findings from studies conducted in patients with long-term exposure to opioids may not generalize to nondependent opioid users. An important implication of this observation is that reinforcing properties of opioid-induced DA release may be an important mechanism in the initiation of opioid use. With prolonged opioid exposure and progression to dependence, other effects of opioids, including effects that are negatively reinforcing, may take on an increasingly dominant role in maintaining use (67).

Methodological factors could also contribute to the divergence of results between studies. First, there are substantial differences in the extent of dopaminergic projections to striatal subdivisions (68). As a consequence, differences in ROI

Table 2. Regional [^{11}C]Raclopride BP_{ND} in the Morphine and Placebo Conditions

	Caudate		Putamen		Nucleus Accumbens		Globus Pallidus (External)	
	Placebo	Morphine	Placebo	Morphine	Placebo	Morphine	Placebo	Morphine
BP_{ND}	3.45 ± 0.5	3.25 ± 0.4	4.11 ± 0.6	3.9 ± 0.4	2.84 ± 0.4	2.60 ± 0.5	2.94 ± 0.5	2.68 ± 0.4

BP_{ND}, nondisplaceable binding potential.

definition could contribute to the divergent findings (69). Second, our study excluded individuals who experienced nausea or marked sedation in response to morphine, which may have enriched the sample for individuals with a high level of opioid reinforcement. Last, differences in the study protocol, specifically the choice of opioid and PET tracer, may have influenced the results. For instance, Spreckelmeyer *et al.* (70) reported that a remifentanil infusion increased striatal DA release measured with [^{18}F]fallypride PET in both abstinent alcohol-dependent individuals and healthy volunteers, with greater magnitudes of the effect than what was observed in this study. Notably, opioids differ in terms of receptor affinity and efficacy (71,72) and abuse liability (73), as well as in their ability to provoke DA release (74,75).

In the present study, we also detected a significant decrease in [^{11}C]raclopride BP_{ND} in response to morphine in the GPe (−9% in BP_{ND}). [^{11}C]Raclopride is commonly used in PET studies to assess $D_{2/3}$ receptor availability and infer DA levels in the striatum. In extrastriatal areas, [^{11}C]raclopride has been considered to be less adequate for measuring DA activity, given the lower density of $D_{2/3}$ receptors in these areas (76). However, the GPe is part of the indirect pathway, wherein neurons express D2 receptors, and there is evidence that decreases in [^{11}C]raclopride BP_{ND} can also be observed in this region, in a range similar to the NAc (77,78). Furthermore, our finding in the GPe remained significant after correction for multiple comparisons and partial volume effects correction, which was performed to account for the limited spatial resolution of the PET camera, which may cause spillover effects in the basal ganglia (79).

In addition to inducing DA release in mesolimbic areas, the dose of morphine administered in our study was sufficient to produce marked objective opioid effects such as miosis and a pronounced subjective response to morphine, including feeling high and feeling drug effects. We explored the possible relationship between subjective effects and changes in [^{11}C]raclopride displacement in response to morphine and noted negative correlations between self-report measures of high and feel drug and ΔBP_{ND} in GPe, and between drug-wanting ratings and ΔBP_{ND} in caudate and putamen. These negative correlations may suggest that the involvement of DA release as measured by [^{11}C]raclopride displacement in modulating opioid-induced reinforcing effects is complex and variable, as previously indicated for several addictive agents (43,80–82). They also support the need to investigate the causal relationship between DA release and opioid reward, for example, by examining whether DA receptor blockade would reduce opioid self-administration in humans.

Our study should be interpreted in light of some limitations. First, our sample size was modest and consisted only of male participants, although it was in line with previous PET studies investigating the effects of different drugs of abuse (9–11,83,84). Also, the sample size did not allow us to investigate whether the *OPRM1* 118G allele carrier status would have conferred a more vigorous DA response to morphine in the ventral striatum, as previously described for alcohol (34). Second, in the ROI analysis, we detected a significant difference in percent change in [^{11}C]raclopride BP_{ND} in the voxels showing the top 10% BP_{ND} values within each ROI. Additionally, after a conservative Bonferroni correction, the ROI-based finding of decreased in [^{11}C]raclopride BP_{ND} in response to morphine in the NAc did not reach significance. Nevertheless, morphine-induced raclopride displacement was identified by two methods, ROI and voxelwise analysis.

In summary, our findings indicate that DA transmission in subcompartments of the human striatum is promoted by morphine. The behavioral significance of this increased DA remains to be established. Future studies, in a larger sample of healthy volunteers as well as in individuals at high risk for OUD, are required to fully delineate the DA contribution to opioid reward and addiction.

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ClinicalTrials.gov: Genetic Effects on Dopamine Response to an Opiate; https://clinicaltrials.gov/ct2/show/NCT01878006; NCT01878006.

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