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Study Importance Questions

What is already known about this subject?

- In people who are lean, dorsal striatal dopamine (DA) release, measured by using positron emission tomography (PET), is greater during fed than fasted conditions.
- Presentation of palatable food without consumption stimulates striatal DA release during overnight fasted conditions in individuals who are lean and in individuals with obesity and binge eating disorder.

What does your study add?

- Viewing high-calorie food images increases striatal DA release when compared with low-calorie food images in people with obesity and prediabetes, but the response is the same during fed and fasted conditions.
- DA release in response to high-calorie food images compared with low-calorie food images is greater in the nucleus accumbens than in the dorsal striatum, suggesting the nucleus accumbens is involved in regulating the motivation to obtain pleasurable reward ("wanting") stimuli.

Abstract

Objective: To determine whether striatal dopamine (DA) release is affected by food ingestion and whether the DA response to high-calorie food images is greater in the fasted than fed state in people with obesity.

Methods: Striatal DA release was evaluated in 10 people with obesity and prediabetes after participants consumed a meal to satiation and after they fasted overnight, and in response to viewing images of high-calorie compared with low-calorie foods after consuming a meal to satiation or fasting overnight, by using positron emission tomography with [^{11}C]raclopride injection.

Results: Striatal DA D2/D3 receptor availability was not different during fasted and fed conditions. Viewing images of high-calorie foods induced striatal DA release relative to viewing images of low-calorie foods ($p<0.05$), but there was no difference in the magnitude of the response between fasting and fed conditions.

Conclusions: People with obesity and prediabetes do not increase striatal DA release after eating a meal to satiation compared with fasting overnight, and fail to inhibit DA release in response to high-calorie food stimuli after eating a meal to satiation. These data suggest impaired DA signaling contributes to greater energy intake during meals in this population.

INTRODUCTION

Obesity is a major public health problem in many countries because of its high prevalence, causal relationship with serious medical comorbidities and economic effects of increased health care costs and work absenteeism (1). In all persons, obesity is caused by ingesting more energy than expended (and needed for organ growth and development in children and adolescents) over a long period of time. This initial positive energy balance generates an accumulation of body fat, which must be maintained by continued high-energy intake.

The brain is the major regulator of energy intake and controls appetite through integration of hedonic and homeostatic mechanisms (2). The neurotransmitter, dopamine (DA), has a central role in regulating reward-motivating behavior with mesolimbic and nigrostriatal dopaminergic signaling in the ventral and dorsal striatum, respectively, required for encoding reward-predictive cues and the motivation to ingest palatable food (3). DA D2/D3 receptor (D2/D3R) availability and DA release can be assessed by using DA signaling-related radioligands in combination with positron emission tomography (PET). By using this technique, an increase in striatal DA release has been observed in people without obesity after consumption of a palatable milkshake (4) or a favorite meal (5) and after presentation of palatable food without consumption (6). In contrast, considerably less is known about DA responses to food cues in people with obesity. Presentation of palatable food without consumption has been shown to stimulate striatal DA release in the overnight fasted state in people with obesity and binge eating disorder (BED) (7) but it is unclear whether feeding modulates this response. In addition, it is currently unknown whether feeding elicits striatal DA release in people with obesity, however, a reduced DA response to glucose ingestion has been reported (8) suggesting the DA response to feeding is altered in people with obesity and insulin resistance.

The main objectives of the present study were to assess striatal DA release in response to consuming a high-calorie meal relative to having fasted overnight and in response to viewing high-calorie food images compared with low-calorie food images during both fasted and fed conditions in people with obesity and insulin resistance, defined by having prediabetes. PET with [^{11}C]raclopride, a nonselective D2/D3R radioligand that is displaced by increased intra-synaptic DA (9), was used to measure striatal D2/D3R availability after eating a meal to satiation compared with overnight fasting conditions and striatal DA release induced by viewing high-calorie compared with low-calorie food images during both fed and fasted conditions. The dorsal and ventral striatal regions were specifically evaluated because these locations contain the highest density of D2/3R in the brain (10) and are involved in reward processing, including viewing and consuming palatable foods (4-6). We hypothesized that D2/D3R availability after the meal would be lower due to meal-induced DA release and DA release in response to the high-calorie food images would be greater during fasted than fed conditions, indicating a modulating effect of hunger on DA release.

METHODS

Study Participants

Ten men (n = 5) and women (n = 5) with obesity completed this study (**Table 1**). Written, informed consent was obtained from all participants before their participation in this study, which was approved by the Human Research Protection Office and the Radioactive Drug Research Committee at Washington University School of Medicine in St. Louis, MO. All participants completed a comprehensive evaluation in the Clinical and Translational Research Unit (CTRU) of Washington University School of Medicine, which included a medical history

and physical examination, standard blood tests, hemoglobin A1c (HbA1c), a 2-h oral glucose tolerance test (OGTT), and dual-energy x-ray absorptiometry scan to assess body fat and fat-free masses (DXA, Lunar iDXA, GE Healthcare Lunar, Madison, WI). Participants were eligible if their body mass index was ≥30 kg/m^2 and they had prediabetes, defined as either HbA1c 5.7-6.4%, fasting plasma glucose concentration of 100-125 mg/dl or 2-h OGTT plasma glucose concentration of 140-199 mg/dl. People with prediabetes were specifically studied to decrease the heterogeneity in insulin sensitivity among participants, which could affect DA signaling (11). Potential participants with a history of diabetes, that smoked more than 10 cigarettes per week, used drugs or medications that can affect DA signaling, consumed excessive amounts of alcohol (>21 units of alcohol per week for men and >14 units of alcohol per week for women), had metal implants that would interfere with the imaging procedures, and were post-menopausal, pregnant or lactating were excluded. All participants were screened for recent drug use by using a urine drug screen (iCup, Redwood Toxicology Laboratory, Santa Rosa, CA). All but one participant tested negative for drug use; this participant tested positive for benzodiazepine use, which was prescribed as needed for anxiety by their primary care physician.

Study Design

Eating behavior characteristics of the participants were assessed by using validated questionnaires that assessed different aspects of eating behavior: i) Questionnaire on Eating and Weight Patterns – Revised (QEWP-R; (12)), Questionnaire on Eating and Weight Patterns – 5 (QEWP-5; (13)) and the Eating Disorder Examination Questionnaire (EDE-Q; (14)) to identify BED; ii) Dutch Eating Behavior Questionnaire (DEBQ; (15)) to assess restrained, emotional, and external eating behavior; iii) Yale Food Addiction Scale (YFAS; (16)) to assess food dependence

by using criteria based on substance dependence diagnosis; and iv) the Food Craving Inventory (FCI; (17)) to assess craving for different types of food.

The study design schema for a single study visit under fasted or fed conditions is shown in **Figure 1**. Each participant completed two PET studies in randomized order, 19 ± 6 days apart. On both occasions, participants fasted for 12 h overnight at home before being admitted to the CTRU where they either continued to fast or consumed a high-calorie meal, self-selected from the hospital cafeteria menu, until satiated. Approximately 30 min after continued fasting or after finishing the meal, participants rated their current level of hunger, satisfaction with their hunger state, fullness, and how much they could eat (scale from 0 to 100). Participants then underwent a brain PET scan (Siemens Biograph mMR, Siemens, Malvern, PA) in the Center for Clinical Imaging Research (CCIR) in conjunction with a bolus intravenous injection of 6-19.4 mCi [^{11}C]raclopride. [^{11}C]raclopride was prepared by using an automated adaptation of published methods (18, 19). Radiochemical purity was >99.5% and specific activity was ≥1104 Ci/mmol (40.8 TBq/mmol). Emission data were collected in 3D mode for 40 frames (30 x 1 min frames; 9 x 2 min frames; 1 x 3 min frame) over 51 min. Structural MR T1-weighted anatomical images were obtained during each PET scan to localize regions of interest (ROI) by using a 3-D Magnetization Prepared Rapid Acquisition Gradient Echo (MP-RAGE) sequence (sagittal orientation, TR = 2300 ms, TE = 2.95 ms, flip angle = 9º, slab thickness = 176 mm, FOV = 240 x 256 mm, voxel dimensions = 1 x 1 x 1 mm) in combination with structural T2-weighted anatomical images (t2_spc_sag_p2_iso, sagittal orientation, TR = 3200 ms, TE = 418 ms, flip angle = 120º, slab thickness = 176 mm, FOV = 256 x 256 mm, voxel dimensions = 1 x 1 x 1 mm) for high resolution segmentation of the cerebellum.

During the first 15 s of the PET scan, participants were shown task instructions. Participants were then shown 96 low-calorie food images for 15 s each over a total of 25 min followed by 96 high-calorie food images for 15 s each over the subsequent 25 min (**Figure 1**). Low-calorie food, rather than non-food, image viewing, was chosen as the 'control' condition to ensure that striatal responsivity to high-calorie food images was due to the high-calorie nature of the images and not to nonspecific features such as simply the act of viewing food images. The presentation of high-calorie food images began at 25 min post-[^{11}C]raclopride injection, consistent with theoretical and validation studies using the single-dose radiotracer method (20-22). A fixation cross was presented for 0.5 s between each food image. Food images (**Table S1)** were selected from a database containing images that have previously been used to examine differences in brain activation responses to high-calorie and low-calorie images (23, 24). High-calorie foods included sweet (e.g., candy, chocolate, cake) and savory (e.g., French fries, burger, pizza) foods, whereas low-calorie foods included vegetables, fruit, and salad. All images had the same resolution and color depth and had the same background color and distance from the camera. E-Prime (Psychology Software Tools, Sharpsberg, PA) software was used to present randomized images within each low-calorie and high-calorie block and collect the participants' responses. During food image presentation, participants rated whether they liked, disliked, or felt neutral towards the thought of eating the food presented by using an E-Prime serial response (SR) box (Psychology Software Tools). Rating results from two participants were excluded from this analysis because the SR box failed to record one participant's responses during their fed study and another participant did not understand the directions during the low-calorie food image portion during their first PET scan.

PET data analyses

Regions of interest (ROIs). The dorsal striatum (whole caudate and whole putamen) and ventral striatum (nucleus accumbens) were identified by using FreeSurfer (25). The dynamic PET images were aligned with each other and co-registered to the individual's MP-RAGE image, as previously described (26). Striatal ROIs and the cerebellar reference region were resampled in the same atlas space (26). The whole cerebellar region was used as the reference region because [^{11}C]raclopride does not accumulate in this region (27) and was segmented by using the spatially unbiased infra-tentorial template (SUIT) toolbox (28) in Matlab R2018b (MathWorks; Natick, MA). Decay-corrected time-activity curves showing radioactivity concentration as a function of time were extracted for each ROI and cerebellum from the dynamic 50 min PET scan. An example time-activity curve and fit to the data using the LSSRM/Bayesian compared to the Logan model (described below) is presented in **Figure 2**.

Assessment of DA D2/D3R Availability, DA Release, and Signal-to-Noise Ratio. To assess DA responses in the fasting and fed states we determined: i) D2/D3R binding potentials (BP_{ND}s) to compare DA release in the fed and fasted states during the entire 50 min PET scan; and ii) the change in displacement of [^{11}C]raclopride from D2/D3R (change in DA release) in response to viewing high-calorie compared with low-calorie food images. BP_{ND}s and the change in DA release in response to viewing high-calorie and low-calorie food images were calculated for each ROI by using a linear extension of the simplified reference region model (LSSRM) (29). A Bayesian method (30) was used to estimate the kinetic parameters, R, k_2, k_{2a}, and γ used in the LSSRM model, in which R is the ratio of transport rates for the binding and reference regions, k_2 represents the clearance of nonspecifically bound radiotracer, k_{2a} represents dissociation from the receptor, and γ measures the extent of displacement of the DA receptor radioligand,

[^{11}C]raclopride, from D2/D3R due to additional synaptic DA released in response to a given stimuli (viewing high-calorie food images) presented during the PET scan (22). D2/D3R BP_{ND} were calculated as k_2/k_{2a} -1 by using dynamic data collected throughout the entire 50 min PET scan. A similar D2/D3R BP_{ND} in both PET scans indicates no difference in DA release over the entire 50 min PET scan in the fasted and fed states, whereas a lower D2/D3R BP_{ND} in the fed state would indicate greater displacement of [^{11}C]raclopride from D2/D3R as a result of food ingestion-induced DA release, as previously shown in people who are lean (5). Our study design does not measure absolute DA release separately during periods of viewing low-calorie and high-calorie food images. However, a delta DA release value can be calculated (γ value) indicating the change in DA release from viewing low-calorie to high-calorie food images (**Figure 2**). For each ROI, the γ value provided an index of DA release in response to viewing images of palatable, high-calorie foods relative to viewing low-calorie food images. γ values were limited to 0 and above. The rate at which transient task-induced DA release returns to baseline, τ, was specified as 0.23 min^{-1}, based on an ROI-derived γ-weighted average τ in 6 initial participants. Signal-to-noise ratio (SNR) was estimated by dividing γ by the standard deviation (SD) of γ.

The LSSRM was specifically developed to analyze time-dependent radioligand displacement during a PET scan allowing alterations in DA release in response to a given stimuli to be modeled. The LSSRM model has been validated and successfully applied to analyze the effect of different reward processing, cognitive and motor tasks during a single PET scan negating the need for multiple PET scans thereby lowering subject burden and radiation exposure (21, 22, 31). In contrast, the commonly-used Logan graphical method (32) does not allow temporal changes in DA release to be resolved requiring separate PET scans to be

performed for each stimuli/intervention studied. For example, if the Logan approach was exclusively used in the present study 4 separate PET scans would have been required (i.e., fasting state viewing low-calorie images, fasting state viewing high-calorie images, fed state viewing low-calorie images and fed state viewing high-calorie images).

Statistical analyses.

Our power calculations were based on previously published changes in DA release in the dorsal striatum in response to meal presentation in people with obesity and BED (7) and in individuals who are lean (6) where Cohen's *d* effect sizes of 1.20 and 1.43 were reported, respectively (6, 7). Using the smaller effect size, a sample size of 10 subjects and 2-tailed tests, we estimated we had sufficient power (>0.90) to detect statistically significant differences in D2/D3R BP_{ND} and DA release in the present study at the ≤0.05 significance level.

Differences in appetitive ratings between fasted and fed states were assessed by using Student's *t*-test for paired samples. A two-way [condition (fasted vs. fed) x type of food image (low- vs. high-calorie) repeated measures analysis of variance (ANOVA) was used to evaluate the effects of feeding and type of food cue on perceived enjoyment of eating the food presented with Tukey's post hoc procedure used to identify significant mean differences. Three-way [brain region (dorsal striatum vs. nucleus accumbens) x condition (fasted vs. fed) x method (LSSRM/Bayesian vs. Logan)] repeated measures ANOVA was used to evaluate the effects of brain region, feeding and method on D2/D3R BP_{ND}. Correlations between LSSRM/Bayesian and Logan-derived BP_{ND} were assessed by using Pearson's product-moment correlation. Two-way [brain region (dorsal striatum vs. nucleus accumbens) x condition (fasted vs. fed)] repeated measures ANOVAs were used to evaluate the effect of viewing palatable, high-calorie food

images on LSSRM/Bayesian-derived γ and SNR values. In addition, separate one-sample *t*-tests were performed to determine whether γ and SNR were greater than 0 for each ROI during fasted and fed conditions. Analyses were conducted using SPSS (Version 25, IBM, Armonk, NY). Data are presented as means ± SEM unless otherwise noted. A p-value < 0.05 was considered statistically significant.

RESULTS

Hunger and pleasure induced by viewing food images

For the study conducted during the fed state, participants consumed 586 ± 106 kcal (16 ± 2% protein, 46% ± 7% carbohydrate, and 38 ± 5% fat) in the morning 30 min before PET scanning was performed. Participants rated themselves as hungrier during the fasted than the fed condition (**Table 2).** In addition, perceived enjoyment of eating was greater when viewing high-calorie than low-calorie food images (main effect of food image type, $p = 0.002$), and when viewing high-calorie foods in the fasted than in the fed state ($p < 0.05$; **Table 2**).

Eating behavior characteristics

Participants reported mild disordered eating behaviors and food cravings but few reported addiction-like eating behavior. However, all participants reported persistent desire or repeated unsuccessful attempts to decrease the consumption of foods high in fat and sugar (**Table 3**). One participant had evidence of BED based on their responses to the QEWP-5. Eliminating this subject from our analyses did not change the statistical significance of any of our findings or conclusions

Striatal D2/D3R availability in the fed and fasted states

Values for D2/D3R BP_{ND} were ~5% higher when using the LSRRM/Bayesian than the Logan graphical method (main effect, $p < 0.001$; **Figure 3A**), and there was a strong correlation between D2/D3R BP_{ND} values obtained using each approach ($r = 0.96$; $p < 0.0001$) (**Figure 3B**). The small difference in D2/D3R BP_{ND} between mathematical models was expected because the Logan graphical method does not include a parameter (γ) representing stimuli-evoked DA release during the PET scan. These data demonstrate the validity of using the LSSRM/Bayesian approach in the current study design to accurately determine BP_{ND} in which DA release during a single scanning period can change when viewing images of high-calorie foods.

Ingestion of a palatable meal did not affect DA release, as there was no difference in D2/D3R BP_{ND} between the fasted and fed states (main effect of condition, $p = 0.91$). However, D2/D3R BP_{ND} was ~30% higher in the dorsal striatum than in the nucleus accumbens (main effect of region, $p < 0.001$) (**Figure 3A**), as expected because of the greater number of D2/D3R in the dorsal striatum (33).

Striatal DA release in response to viewing high-calorie food images

Displacement of the DA receptor radioligand [^{11}C]raclopride from D2/D3R due to additional synaptic DA release (γ) in response to viewing images of high-calorie foods relative to viewing low-calorie foods was greater than zero in the dorsal striatum and nucleus accumbens during both fasted and fed conditions ($p < 0.05$ for all comparisons; **Figure 3C**). However, the extent of synaptic DA release during the fasting condition was not different than during the fed condition ($p = 0.64$), but was 2-4-fold greater in the nucleus accumbens than in the dorsal striatum ($p < 0.01$). ROI volume is not expected to affect γ because γ measures the size of the

decrease in [^{11}C]raclopride signal when stimuli change from low-calorie to high-calorie food images. The SNR for γ was also significantly greater than 0 in all conditions ($p < 0.01$ for all comparisons), but did not differ between fasted and fed conditions ($p = 0.43$) or between dorsal striatum and nucleus accumbens ($p = 0.11$) (**Figure 3D**).

Discussion

Non-homeostatic food intake cued by high-calorie food stimuli is regulated, in part, by central DA signaling (3). In the present study, we used brain PET in conjunction with the injection of [^{11}C]raclopride, a displaceable nonselective D2/D3R radioligand, to measure striatal D2/D3R availability and DA release in people with obesity and prediabetes who viewed palatable, high-calorie food images during fasted and fed conditions. In contrast to previous observations in people who are not obese (5), we found meal ingestion did not affect striatal D2/D3R availability, indicating no effect on DA release relative to the fasting state, and that striatal DA release in response to viewing high-calorie relative to low-calorie food images was similar in the fasted and fed states in people with both obesity and prediabetes. These findings suggest reward region responsivity to high-calorie food ingestion is not adequately regulated in people with both obesity and prediabetes, thereby contributing to greater energy intake with meals.

Few studies have evaluated the potential importance of food intake and viewing food images on brain activation and DA signaling in people. Moreover, we are not aware of any PET studies that used [^{11}C]raclopride to evaluate DA release in people with obesity in both fasted and fed states. In people that are not obese, striatal activation in response to high-calorie food images, assessed by using fMRI, was greater in the fasted than in the fed state (34), and striatal DA

release, assessed by using PET with [^{11}C]raclopride, was greater after consuming a palatable milkshake (4) or a favorite meal (5) and after presenting palatable food without consumption (6). In people with obesity, striatal activation, assessed by using fMRI, is increased in the fasting state in response to viewing high-calorie food images, and this activation was greater than that observed in people who are lean (35, 36). Our study extends these findings by demonstrating no difference in DA release between the fed and fasted states in contrast to previous findings in people that were not obese (5). Although we found an increase in striatal DA release in response to viewing high-calorie relative to low-calorie food images in people with obesity with prediabetes, the DA response was the same during fed and fasted conditions. Our findings and those of these previous fMRI and PET studies support the dynamic vulnerability and incentive sensitization hypotheses of obesity (37), which posit that persistent overeating of high-calorie foods reduces reward region responsivity to high-calorie food ingestion and instead results in increased incentive salience ("wanting") prompted by conditioned food cues. The mechanism(s) responsible for altered striatal DA signaling in obesity is not known, but is possibly related to: i) changes in the levels of hypothalamic neuropeptides involved in energy homeostasis, such as leptin and ghrelin, which directly modulate the activity of dopaminergic neurons (38); ii) increased central insulin concentrations that act directly on the DA system (39); iii) altered glucose metabolism (40); and/or iv) greater catecholaminergic afferents arising from the hindbrain (41).

We found DA release in response to high-calorie food images was greater in the nucleus accumbens (located in the ventral striatum) than in the dorsal striatum, which is analogous to responses to other rewarding stimuli, including alcohol (42), monetary reward (43) and amphetamine administration (44). Increased DA release in nucleus accumbens relative to dorsal

striatum may be due to the functional importance of the former in incentive salience, which involves the motivation to obtain a pleasurable reward ("wanting") (3). In contrast, the dorsal striatum plays a larger role in "action salience" (45), which increases the motivation to carry out the motor actions necessary to obtain the pleasurable reward. Our participants might have experienced "wanting" during exposure to high-calorie food images with less urge to act to obtain food, because there was no expectation that food would be consumed during the PET scan.

Our study has several limitations. We purposely studied people with obesity who had prediabetes to reduce the heterogeneity in insulin sensitivity among participants, which could affect DA signaling (11) and make it more difficult to detect differences in fed and fasted conditions. It is therefore possible that our results might not translate to people with obesity and normal oral glucose tolerance. In addition, our study was conducted in a small number of participants, which could have failed to detect a difference between fed and fasted states and we did not evaluate DA signaling in a healthy comparator lean group. However, our *a priori* power calculations found the number of participants in our study (n=10) would be sufficient to detect a meaningful difference in DA release between the fasting and fed states that had been shown previously in people who were not obese. Moreover, the number of subjects included in the present study is greater than the number of subjects in a previous study (n=7) that reported an increase in striatal DA release in people without obesity after they consumed a favorite meal (5).

In conclusion, we found striatal DA release after eating to satiety was not different than DA release after fasting for 12-hour overnight in people with obesity and prediabetes. These results differ from previous observations of greater striatal DA release in the fed than fasted state in people who are not obese (5), and suggest altered DA signaling contributes to greater energy

intake and sensitivity to food cues after initiating food consumption in people with obesity and prediabetes than those who are lean.

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References

1. Klein S, Wadden T, Sugerman HJ. AGA technical review on obesity. Gastroenterology. 2002;123(3):882-932.
2. Berthoud HR, Munzberg H, Morrison CD. Blaming the Brain for Obesity: Integration of Hedonic and Homeostatic Mechanisms. Gastroenterology. 2017;152(7):1728-38.
3. Berridge KC, Ho CY, Richard JM, DiFeliceantonio AG. The tempted brain eats: pleasure and desire circuits in obesity and eating disorders. Brain research. 2010;1350:43-64.
4. Thanarajah SE, Backes H, DiFeliceantonio AG, et al. Food Intake Recruits Orosensory and Post-ingestive Dopaminergic Circuits to Affect Eating Desire in Humans. Cell Metab. 2019;29(3):695-706 e4.
5. Small DM, Jones-Gotman M, Dagher A. Feeding-induced dopamine release in dorsal striatum correlates with meal pleasantness ratings in healthy human volunteers. Neuroimage. 2003;19(4):1709-15.
6. Volkow ND, Wang GJ, Fowler JS, et al. "Nonhedonic" food motivation in humans involves dopamine in the dorsal striatum and methylphenidate amplifies this effect. Synapse. 2002;44(3):175-80.
7. Wang GJ, Geliebter A, Volkow ND, et al. Enhanced striatal dopamine release during food stimulation in binge eating disorder. Obesity (Silver Spring, Md). 2011;19(8):1601-8.
8. Wang GJ, Tomasi D, Convit A, et al. BMI modulates calorie-dependent dopamine changes in accumbens from glucose intake. PLoS One. 2014;9(7):e101585.

9. Dewey SL, Smith GS, Logan J, Brodie JD, Fowler JS, Wolf AP. Striatal binding of the PET ligand 11C-raclopride is altered by drugs that modify synaptic dopamine levels. Synapse. 1993;13(4):350-6.

10. Siessmeier T, Zhou Y, Buchholz HG, et al. Parametric mapping of binding in human brain of D2 receptor ligands of different affinities. J Nucl Med. 2005;46(6):964-72.

11. Volkow ND, Wang GJ, Baler RD. Reward, dopamine and the control of food intake: implications for obesity. Trends Cogn Sci. 2011;15(1):37-46.

12. Spitzer RL, Yanovski SZ, Marcus MD. Questionnaire on Eating and Weight Patterns - Revised. Pittsburgh, PA: Behavioral Measurement Database Services; 1994.

13. Yanovski SZ, Marcus MD, Wadden TA, Walsh BT. The Questionnaire on Eating and Weight Patterns-5: an updated screening instrument for binge eating disorder. Int J Eat Disord. 2015;48(3):259-61.

14. Fairburn CG, Beglin SJ. Assessment of eating disorders: interview or self-report questionnaire? Int J Eat Disord. 1994;16(4):363-70.

15. van Strien T, Frijters JER, Bergers GPA, Defares PB. The Dutch Eating Behavior Questionnaire (DEBQ) for Assessment of Restrained, Emotional, and External Eating Behavior. Int J Eat Disord. 1986;5(2):295-315.

16. Gearhardt AN, Corbin WR, Brownell KD. Preliminary validation of the Yale Food Addiction Scale. Appetite. 2009;52(2):430-6.

17. White MA, Whisenhunt BL, Williamson DA, Greenway FL, Netemeyer RG. Development and validation of the food-craving inventory. Obes Res. 2002;10(2):107-14.

18. Ehrin E, Gawell L, Hogberg T, Depaulis T, Strom P. Synthesis of [Methoxy-H-3]- and [Methoxy-C-11]- Labeled Raclopride - Specific Dopamine-D2 Receptor Ligands. J Labelled Compd Rad. 1987;24(8):931-40.

19. Farde L, Pauli S, Hall H, et al. Stereoselective binding of 11C-raclopride in living human brain--a search for extrastriatal central D2-dopamine receptors by PET. Psychopharmacology (Berl). 1988;94(4):471-8.

20. Badgaiyan RD, Fischman AJ, Alpert NM. Striatal dopamine release during unrewarded motor task in human volunteers. Neuroreport. 2003;14(11):1421-4.

21. Badgaiyan RD, Fischman AJ, Alpert NM. Striatal dopamine release in sequential learning. Neuroimage. 2007;38(3):549-56.

22. Alpert NM, Badgaiyan RD, Livni E, Fischman AJ. A novel method for noninvasive detection of neuromodulatory changes in specific neurotransmitter systems. Neuroimage. 2003;19(3):1049-60.

23. Blechert J, Meule A, Busch NA, Ohla K. Food-pics: an image database for experimental research on eating and appetite. Front Psychol. 2014;5:617.

24. Ulrich M, Steigleder L, Gron G. Neural signature of the Food Craving Questionnaire (FCQ)-Trait. Appetite. 2016;107:303-10.

25. Fischl B, Salat DH, Busa E, et al. Whole brain segmentation: automated labeling of neuroanatomical structures in the human brain. Neuron. 2002;33(3):341-55.

26. Eisenstein SA, Koller JM, Piccirillo M, et al. Characterization of extrastriatal D2 in vivo specific binding of [(1)(8)F](N-methyl)benperidol using PET. Synapse. 2012;66(9):770-80.

27. Farde L, Halldin C, Stone-Elander S, Sedvall G. PET analysis of human dopamine receptor subtypes using 11C-SCH 23390 and 11C-raclopride. Psychopharmacology (Berl). 1987;92(3):278-84.

28. Diedrichsen J, Balsters JH, Flavell J, Cussans E, Ramnani N. A probabilistic MR atlas of the human cerebellum. Neuroimage. 2009;46(1):39-46.

29. Friston KJ, Malizia AL, Wilson S, Cunningham VJ, Jones T, Nutt DJ. Analysis of dynamic radioligand displacement or "activation" studies. J Cereb Blood Flow Metab. 1997;17(1):80-93.

30. Bretthorst GL. Bayesian Data-Analysis Toolbox. 4.21 ed2014.

31. Ceccarini J, Vrieze E, Koole M, et al. Optimized in vivo detection of dopamine release using 18F-fallypride PET. J Nucl Med. 2012;53(10):1565-72.

32. Logan J. Graphical analysis of PET data applied to reversible and irreversible tracers. Nucl Med Biol. 2000;27(7):661-70.

33. Mawlawi O, Martinez D, Slifstein M, et al. Imaging human mesolimbic dopamine transmission with positron emission tomography: I. Accuracy and precision of D(2) receptor parameter measurements in ventral striatum. J Cereb Blood Flow Metab. 2001;21(9):1034-57.

34. Goldstone AP, Prechtl de Hernandez CG, Beaver JD, et al. Fasting biases brain reward systems towards high-calorie foods. Eur J Neurosci. 2009;30(8):1625-35.

35. Stoeckel LE, Weller RE, Cook EW, 3rd, Twieg DB, Knowlton RC, Cox JE. Widespread reward-system activation in obese women in response to pictures of high-calorie foods. Neuroimage. 2008;41(2):636-47.

36. Rothemund Y, Preuschhof C, Bohner G, et al. Differential activation of the dorsal striatum by high-calorie visual food stimuli in obese individuals. Neuroimage. 2007;37(2):410-21.

37. Stice E, Burger K. Neural vulnerability factors for obesity. Clin Psychol Rev. 2019;68:38-53.

38. Kenny PJ. Common cellular and molecular mechanisms in obesity and drug addiction. Nat Rev Neurosci. 2011;12(11):638-51.

39. Berthoud HR. Metabolic and hedonic drives in the neural control of appetite: who is the boss? Curr Opin Neurobiol. 2011;21(6):888-96.

40. de Araujo IE, Lin T, Veldhuizen MG, Small DM. Metabolic regulation of brain response to food cues. Curr Biol. 2013;23(10):878-83.

41. Mejias-Aponte CA, Drouin C, Aston-Jones G. Adrenergic and noradrenergic innervation of the midbrain ventral tegmental area and retrorubral field: prominent inputs from medullary homeostatic centers. J Neurosci. 2009;29(11):3613-26.

42. Boileau I, Assaad JM, Pihl RO, et al. Alcohol promotes dopamine release in the human nucleus accumbens. Synapse. 2003;49(4):226-31.

43. Martin-Soelch C, Szczepanik J, Nugent A, et al. Lateralization and gender differences in the dopaminergic response to unpredictable reward in the human ventral striatum. Eur J Neurosci. 2011;33(9):1706-15.

44. Martinez D, Slifstein M, Broft A, et al. 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. 2003;23(3):285-300.

45. Berridge KC, Robinson TE, Aldridge JW. Dissecting components of reward: 'liking', 'wanting', and learning. Curr Opin Pharmacol. 2009;9(1):65-73.

Figure legends

Figure 1. Schematic illustration of the study visit. Participants completed 2 PET scans performed in a randomized order. On each occasion participants arrived in the Research Unit in the fasting state then during the hour before each positron emission tomography/magnetic resonance (PET/MR) scan with [^{11}C]raclopride injection they consumed a palatable meal or remained in the fasting state and completed appetitive ratings. During the PET/MR scan, participants rated images of low-calorie and high-calorie foods shown during the first and last 25 min of the scan, respectively. Fixation crosses were shown for 0.5 s after each food image. Examples of low- and high-calorie food images are shown.

Figure 2. Representative time activity curve (TAC) and fit to the [^{11}C]raclopride radioactive signal in the nucleus accumbens for a single participant. The TAC in the cerebellar region of the same participant was used as the reference TAC because [^{11}C]raclopride does not accumulate in this region. To assess DA responses in the fasting and fed states we determined: i) D2/D3R binding potentials (BP_{ND}s) to compare DA release in the fed and fasted states during the entire 50 min PET scan; and ii) the change in displacement of [^{11}C]raclopride from D2/D3R (change in DA release) in response to viewing high-calorie compared with low-calorie food images. D2/D3R BP_{ND}s were calculated based on the distribution volume ratio (DVR) with the distribution volume in each region of interest (ROI) related to the number of unoccupied binding sites and the DVR the ratio of the DV in a receptor-containing region (i.e., nucleus accumbens or dorsal striatum) to the cerebellum (i.e., the reference nonreceptor region). The vertical lines at 25 min indicate the end of the low-calorie food image condition and the start of the high-calorie

food image condition. γ indicates [^{11}C]raclopride displacement by endogenous dopamine release in response to viewing high-calorie relative to low-calorie food images. While γ is estimated using data from the entire positron emission tomography (PET) scan, the biggest effect on γ is due to the first few minutes after change from low-calorie to high-calorie food image condition since the linear extension of the simplified reference region model (LSSRM) assumes diminishing effect of changed conditions on dopamine release. The solid line indicates [^{11}C]raclopride signal as predicted by a model in which $\gamma = 0$ (i.e., no high-calorie image-induced dopamine release).

Figure 3. (**A**) Effect of consuming a meal to satiation on striatal dopamine D2/D3 receptor binding potential (D2/D3R BP_{ND}) assessed by using linear extension of the simplified reference region (LSSRM)/Bayesian and Logan graphical (Logan) models. (**B**) Relationship between D2/D3R BP_{ND} values obtained by using the LSRRM/Bayesian and the Logan graphical methods. Circles and triangles represent values for dorsal striatum and nucleus accumbens, respectively. **(C)** Striatal dopamine release (γ), estimated by using the LSSRM/Bayesian method, in response to viewing high-calorie compared with low-calorie food images during fed and fasted conditions. **(D)** Effect of feeding condition on signal-to-noise ratio of γ (SNR; γ/(SD of γ), estimated by the LSSRM/Bayesian method. Data are means ± SEM in panels **A**, **C** and **D**. *Values different from nucleus accumbens, $p < 0.01$. $^{\#,\#\#}$ Values greater than zero, $p \leq 0.02, 0.01$.

Table 1. Characteristics of the study participants.

Age (yrs)	42 ± 3
Body mass index (kg/m^2)	39.4 ± 1.5
Body fat (%)	46.8 ± 2.0
Fasting glucose (mg/dl)	99 ± 4
2-h OGTT glucose (mg/dl)	162 ± 7
Fasting insulin (μU/ml)	23 ± 3
HbA1c (%)	5.5 ± 0.1
Triglyceride (mg/dl)	144 ± 17
HDL-cholesterol (mg/dl)	43 ± 3

Data are means ± SEM. Abbreviations: OGTT, oral glucose tolerance test; HbA1c, hemoglobin A1c; HDL, high-density lipoprotein.

Table 2. Appetitive ratings and perceived enjoyment of eating low-calorie and high-calorie foods in the fasted and fed states.

	Fasted	Fed
Appetitive ratings immediately before the PET study		
How hungry are you?	45.7 ± 9.0	13.2 ± 5.1*
How full are you?	28.5 ± 8.7	67.6 ± 9.4*
How satisfied are you?	33.1 ± 8.5	79.0 ± 3.5*
How much can you eat?	58.4 ± 7.2	22.2 ± 5.2*
Perceived enjoyment during the PET study		
Eating low-calorie foods	1.52 ± 0.08	1.51 ± 0.07
Eating high-calorie foods	1.15 ± 0.02†	1.24 ± 0.05*†

Data are means ± SEM. Appetitive ratings ranged from 0-100. Perceived enjoyment scores ranged from 1-3 with lower scores indicate greater "liking" of imagining eating the food depicted in the presented image. * Value significantly different from corresponding fasted value, $p < 0.05$. † Value significantly different from corresponding low-calorie food value, $p < 0.05$.

Table 3. Eating behavior characteristics

Dutch Eating Behavior Questionnaire	
Restrained Eating Scale	2.3 ± 0.2
External Eating Scale	3.0 ± 0.1
Emotional Eating Scale	2.2 ± 0.2
Clearly Labeled Emotions Scale	2.2 ± 0.2
Diffuse Emotions Scale	2.5 ± 0.2
Yale Food Addiction Scale	
Symptom counts	2.1 ± 0.4
Substance taken in larger amount and for longer period than intended	1/10
Persistent desire or repeated unsuccessful attempts to quit	10/10
Much time/activity to obtain, use, recover	2/10
Important social, occupational, or recreational activities given up or reduced	0/10
Use continues despite knowledge of adverse consequences	4/10
Tolerance	3/10
Characteristic withdrawal symptoms; substance taken to relieve withdrawal	0/10
Use causes clinically significant impairment or distress	1/10
Food Dependence Diagnosis	1/10
Food Craving Inventory	
Fat	2.2 ± 0.2
Sweet	2.5 ± 0.2
Carbohydrate	2.1 ± 0.2
Fast-food Fat	2.9 ± 0.2

Data are means ± SEM or frequency of occurrence. The Dutch Eating Behavior Questionnaire scores range from 1 (never) – 5 (very often) on each scale. Yale Food Addiction Scale symptom counts range from 0-8. Food Craving Inventory scores range from 1 (not at all) – 5 (nearly every day) for each food type.