

Satiation attenuates BOLD activity in brain regions involved in reward and increases activity in dorsolateral prefrontal cortex: an fMRI study in healthy volunteers^{1–4}

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ABSTRACT

Background: Neural responses to rewarding food cues are significantly different in the fed vs. fasted (>8 h food-deprived) state. However, the effect of eating to satiety after a shorter (more natural) intermeal interval on neural responses to both rewarding and aversive cues has not been examined.

Objective: With the use of a novel functional magnetic resonance imaging (fMRI) task, we investigated the effect of satiation on neural responses to both rewarding and aversive food tastes and pictures.

Design: Sixteen healthy participants (8 men, 8 women) were scanned on 2 separate test days, before and after eating a meal to satiation or after not eating for 4 h (satiated vs. premeal). fMRI blood oxygen level-dependent (BOLD) signals to the sight and/or taste of the stimuli were recorded.

Results: A whole-brain cluster-corrected analysis ($P < 0.05$) showed that satiation attenuated the BOLD response to both stimulus types in the ventromedial prefrontal cortex (vmPFC), orbitofrontal cortex, nucleus accumbens, hypothalamus, and insula but increased BOLD activity in the dorsolateral prefrontal cortex (dlPFC; local maxima corrected to $P \leq 0.001$). A psychophysiological interaction analysis showed that the vmPFC was more highly connected to the dlPFC when individuals were exposed to food stimuli when satiated than when not satiated.

Conclusions: These results suggest that natural satiation attenuates activity in reward-related brain regions and increases activity in the dlPFC, which may reflect a “top down” cognitive influence on satiation. This trial was registered at clinicaltrials.gov as NCT02298049. *Am J Clin Nutr* 2015;101:697–704.

Keywords: fMRI, hypothalamus, prefrontal cortex, reward, satiation

INTRODUCTION

The frequency and size of meals are influenced by activity in brain circuitry that processes nutritional state signals and food reward value (1). Thus, hunger associated with food deprivation increases the incentive value of food, which is reflected in enhanced responses to appetitive stimuli in reward-related brain areas, as assessed by fMRI (2–6), whereas the consumption of food is associated with reduced activity in reward circuitry (7).

A role for frontal neural circuitry in the control of food intake was also highlighted recently (8). Dorsolateral prefrontal cortex (dlPFC)⁵ activation has been associated with higher levels of

self-control over food choices and cognitive restraint of intake (9, 10). This raises the possibility that the reduced motivation to eat associated with satiation is mediated in part by enhanced activity in prefrontal brain regions important for higher cognitive functions and decision making. Activity in the dlPFC may affect food motivation by modulating reward value signals encoded by the ventromedial prefrontal cortex (vmPFC) (9, 11), suggesting that interactions between the dlPFC and vmPFC may be important in satiation, although this remains to be investigated.

Previous fMRI studies focused on the effects of prolonged fasting on brain responses to food-related stimuli (12–14) or on sensory-specific responses rather than the effects of satiation per se (15–17). Hence, the results from previous work in “hungry” participants may not reflect typical satiety processes. To our knowledge, no study has assessed the effects of a meal on neural responses to food cues and compared this with a condition simulating natural intermeal hunger levels. In addition, there is some evidence to suggest that the ingestion of food leads to a selective decline in positive hedonic reactions with no effect on aversive reactions (18). However, there has been no investigation to date of the effects of satiation on neural responses

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³ Supplemental Table 1 is available from the “Supplemental data” link in the online posting of the article and from the same link in the online table of contents at <http://ajcn.nutrition.org>.

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⁵ Abbreviations used: BDI, Beck Depression Inventory; dlPFC, dorsolateral prefrontal cortex; FMRI, the Oxford Centre for Functional Magnetic Resonance Imaging of the Brain; FWE, family-wise error; PFC, prefrontal cortex; STAI, State-Trait Anxiety Inventory; VAS, visual analog scale; vmPFC, ventromedial prefrontal cortex.

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to both rewarding and aversive food-related stimuli in the same study. Furthermore, previous studies usually only assessed responses to either taste or visual stimuli and thus were unable to investigate modality-specific effects of satiation on neural responses. We developed an fMRI paradigm that reliably activates neural responses to both primary (tastes) and secondary (pictures) rewarding and aversive appetitive stimuli in the human brain (19). In the present study, we assessed the effects of natural satiety on neural and behavioral responses to these stimuli.

We predicted that, compared with a premeal state, consuming a satiating lunch would reduce BOLD activity to rewarding stimuli in areas such as the vmPFC, orbitofrontal cortex, ventral striatum, hypothalamus, insula, amygdala, and hippocampus. We also predicted that satiation would increase BOLD responses to rewarding stimuli in the dlPFC, which would be negatively correlated with activity in the vmPFC, based on recent research showing an inverse relation between these 2 areas in a food task (9). We predicted that satiation would have no effect on neural response to aversive stimuli.

METHODS

Subjects

Sixteen healthy volunteers [8 men, 8 women; mean \pm SE age: 21.7 ± 0.9 y; mean \pm SE BMI (in kg/m^2): 21.1 ± 0.4] who met the inclusion criteria were recruited via posters and local advertisements on websites. The study was advertised as a chocolate experiment during fMRI scanning, with a free lunch of cheese sandwiches and £50 compensation. Ethics approval was provided by the Oxford Research Ethics Committee B (National Research Ethics Service), and informed consent was obtained from all participants. The study was conducted in accordance with the ethical standards laid down in the 1975 Declaration of Helsinki, as revised in 1983. Participants were excluded if they had a past or present Axis I disorder, as determined by the Structured Clinical Interview for DSM Disorders (20), or a score >10 on the Beck Depression Inventory (BDI) (21). Other exclusion criteria were taking medication, being left-handed, being a smoker, having food allergies, having diabetes, being aged <18 or >60 y, having a BMI outside the range of 18.5–24.9, or having any contraindications to fMRI scanning such as the presence of a pacemaker.

Design

A within-subjects design was used, with participants taking part in both experimental conditions: premeal and satiated. For both conditions (each conducted on a separate test day), participants were scanned twice, 1 h apart. In the satiated condition, participants received lunch between the scans, whereas those in

the premeal condition received lunch after the second scan of the test day and had a 1-h break between the scans (**Figure 1**). Baseline scans were acquired so that they could be subtracted from subsequent scans during analysis (e.g., satiated scan minus baseline scan from the same test day) to control for baseline differences between test days. Participants were tested within a month of screening, and test days were either 1 or 2 wk apart, with scans at the same time and on the same day of the week as the first sessions (± 30 min). The order of completing the satiated and premeal test days was counterbalanced across participants and sexes.

Scanner stimuli and task

The scanner stimuli and task are described in full elsewhere (19, 22). In brief, the taste stimuli comprised a tasteless rinse control, a chocolate taste, and an unpleasant strawberry taste (all were liquid at room temperature). The tastes were delivered via polytetrafluoroethylene tubes in 0.5-mL bursts while participants were in the scanner. Images shown in the scanner consisted of a gray control image, an image of chocolate, and an image of moldy strawberries. The scanner task comprised 6 conditions (presented 9 times): chocolate taste, chocolate picture, chocolate taste with chocolate picture, strawberry taste, strawberry picture, and strawberry taste with strawberry picture. Each presentation lasted 7 s. Participants also made subjective ratings after each stimulus exposure (rated pleasantness, intensity, and wanting) using a button box.

Lunch

The lunch consisted of a glass of water and an ad libitum meal of cheddar cheese sandwiches on oatmeal bread. A single sandwich weighed ~ 120 g (358 calories), and participants were able to eat a maximum of 8 sandwiches (~ 960 g). Four sandwiches were initially provided to each participant, and participants were given free access to additional sandwiches if they wished to consume more. The use of this bland savory food ensured that participants were satiated while minimizing sensory-specific satiety effects, because the tested stimuli were sweet and distinct from the sandwiches (15).

Procedure

Screening day

Participants were screened with the Medical Screening Sheet, the Structured Clinical Interview for DSM Disorders, and the BDI to determine eligibility. They were also given the Fawcett-Clark Pleasure Scale to examine their capacity for pleasure (23), the Snaith-Hamilton Pleasure Scale as an indicator of anhedonia (24), the Eating Attitudes Test questionnaire as a measure of disordered eating (25), and the Three-Factor Eating Questionnaire to examine eating style (26). Participants also completed an

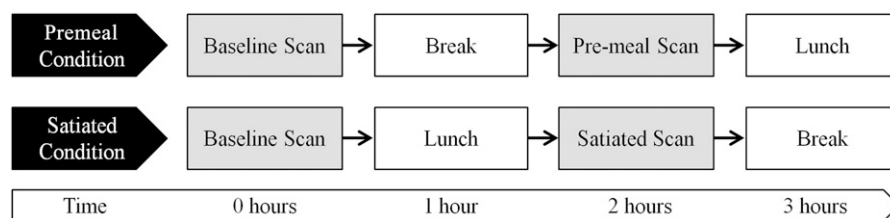


FIGURE 1 Overview of experimental procedure: premeal condition and satiated condition with approximate timings.

fMRI screening sheet to ensure they were eligible to take part in the study. Their height and weight were measured to allow calculation of BMI, and they completed a trial run of the scanner task to ensure they liked and disliked the chocolate and strawberry liquids, respectively (ensuring participants made positive ratings of chocolate stimuli and negative ratings of strawberry stimuli). Participants also ate a sample of the cheese sandwiches to ensure that they would be willing to eat the same sandwiches on the test days. A sandwich questionnaire was used to assess liking of the cheese sandwich; the questionnaire used a 100-mm visual analog scale (VAS) and asked participants to rate the sandwich on how enjoyable it was and whether they would choose to eat it. A chocolate questionnaire was also used to assess chocolate craving, liking, frequency of consumption, and amount consumed (27).

Testing day

Participants were asked not to eat chocolate for 24 h before the scan, which was checked verbally on the morning of the test. They were also asked not to eat food for at least 2 h before scanning to ensure a premeal state, which was checked by having participants complete a breakfast questionnaire detailing when they last ate. Scanning took place either between 1130 and 1430 h or 1230 and 1530 h. On arrival, participants filled out a breakfast form, the State-Trait Anxiety Inventory (STAI) as a measure of anxiety (28), and VASs before completing the first scan of the day. Each VAS questionnaire used 100-mm scales and contained mood-related items (alertness, disgust, drowsiness, anxiety, happiness, nausea, sadness, withdrawn, and faint) and appetite-related items (hunger, fullness, desire to eat, and thirst). After this, participants completed another set of appetite VASs. Participants in the satiated condition were then invited to consume their lunch and asked to eat to the nearest half sandwich to facilitate estimating the amount of food eaten (either at 1230 or 1330 h, depending on the starting time). After they had finished eating, participants were asked to fill in a sandwich rating form and appetite VAS before completing the second scan. After the second scan, participants completed a final set of appetite and mood VASs and the STAI. Participants in the premeal condition completed an appetite VAS after the first scan and were then given a 1-h break outside the scanner (instead of eating lunch). During this time, participants were allowed to read a book or a newspaper in an adjacent room. This break was followed by an appetite VAS and the second scan. Subsequently, participants were given an appetite VAS and lunch (as above, with the same sandwich rating) before completing a final set of mood and appetite VASs along with the STAI. Participants then returned for a second scanning day and underwent the entire procedure again for the condition they did not complete the first time.

fMRI scan

An event-related interleaved design was used that utilized the 6 scanner task stimuli described above in random permuted sequence. A 3.0-T Magnetom Verio (Siemens) whole-body scanner was used at the Oxford Centre for Functional Magnetic Resonance Imaging of the Brain (FMRIB), acquiring T2-weighted echo planar imaging slices every 2 s (TR = 2). Thirty-six axial slices (in-plane resolution of $3 \times 3 \times 3$ mm, no gap) were acquired, the matrix size was 64×64 mm, and the field of view

was 192×192 mm. Acquisition took place during the task, resulting in a total of 976 volumes (the first 4 being dummy scans). A whole-brain T2-weighted echo planar imaging volume of these dimensions and an anatomic T1 volume with axial plane slice thickness of 1 mm and in-plane resolution of $1.0 \times 1.0 \times 1.0$ mm were also obtained.

fMRI analysis

The FMRIB software library (www.fmrib.ox.ac.uk/fsl) was used for preprocessing and statistical analyses of the data. For preprocessing, the following were used: high-pass filter cutoff of 60 s, motion correction using FMRIB's Linear Image Registration Tool (MCFLIRT), interleaved slice timing correction, spatial smoothing with a 6-mm full-width-half-maximum kernel, high-pass temporal filtering, and film prewhitening. Functional data were registered to their corresponding structural images and transformed to Montreal Neurological Institute space with the use of a reference brain (12 df linear transformation). Multivariate Exploratory Linear Optimized Decomposition into Independent Components (MELODIC) was also used to remove artifacts (mean \pm SE components removed: $4.98 \pm 0.27\%$).

After this, the MELODIC filtered scans were entered into a first-level analysis to produce contrasts of the 6 experimental stimuli from the task minus the corresponding control stimuli (control rinse, control gray image, and combination of both). Motion correction variables for each of the 6 experimental stimuli were entered as regressors of no interest and orthogonalized to the total of 9 experimental and control stimuli.

To account for baseline, a second-level analysis (fixed effects) was conducted on these first-level contrasts for each participant to subtract baseline scans from the postmanipulation scans (i.e., premeal minus baseline and satiated minus baseline). Subsequently, contrasts produced at the second level were entered into a third-level mixed-effects (FMRIB's Local Analysis of Mixed Effects 1+2) group analysis: premeal (baseline subtracted) minus satiated (baseline subtracted). *F* tests were produced for the following main effects and interactions: condition, stimuli, modality, condition \times stimuli, condition \times modality, stimuli \times modality, and condition \times stimuli \times modality. By using a backward elimination process, nonsignificant variables were removed one at a time (interactions first, followed by main effects) until only variables that were significant remained.

Group *Z* statistic images were subsequently corrected for multiple comparisons by means of family-wise error (FWE) correction to control for false positives with the use of the AlphaSim program, which is part of the AFNI toolkit (29). To control the FWE rate, the program takes the particular voxel-wise threshold, voxel dimensions, and spatial smoothing kernel size used in the fMRI analysis and, by means of a Monte Carlo simulation, computes the probability of a cluster of specific size arising by chance. On the basis of our data, with a voxel-wise threshold of $P < 0.001$ (z score > 3.1) only clusters with > 24 contiguous voxels were significant with an FWE-corrected $P < 0.05$. Because the hypothalamus is difficult to image accurately because of its proximity to sinuses and susceptibility to artifacts (30), and is also a particularly small region yielding small cluster sizes in other appetite-fMRI studies (31), a more conservative voxel-wise threshold ($P < 0.0005$, z score > 3.3) producing a smaller FWE cluster threshold (≥ 19 contiguous

TABLE 1
Participant characteristics¹

Measure	Baseline value
TFEQ	
Restraint Scale	3.8 ± 0.8
Disinhibition Scale	4.6 ± 0.6
Hunger Scale	3.9 ± 0.8
STAI	
State	30.1 ± 1.7
Trait	30.4 ± 1.7
FCPS	131.5 ± 3.7
SHAPS	22.3 ± 1.0
BDI	0.8 ± 0.4
EAT	2.4 ± 0.5
Chocolate liking	8.4 ± 0.3
Chocolate craving	6.1 ± 0.4
Premeal condition intake, g	336.9 ± 40.5
Satiated condition intake, g	398.1 ± 41.8

¹Values are means ± SEMs; $n = 16$. Data were analyzed with paired t tests where appropriate. BDI, Beck Depression Inventory; EAT, Eating Attitudes Test questionnaire; FCPS, Fawcett-Clark Pleasure Scale; SHAPS, Snaith-Hamilton Pleasure Scale; STAI, State-Trait Anxiety Inventory; TFEQ, Three-Factor Eating Questionnaire.

voxels; $P < 0.05$) was used to balance the risk of type 1 vs. type 2 errors. Effects of stimuli and modality were not reported unless they interacted with satiation. For clarity, local maxima are reported in the tables of activation results.

Psychophysiological interaction analysis

A psychophysiological interaction analysis was used to identify whether satiation was associated with enhanced connectivity between the dlPFC and the vmPFC. For each participant, a 3-mm radius sphere was placed at the individual peak dlPFC voxel for the effect of condition within the group-level mask of the dlPFC. The BOLD first eigen-time series was then extracted for each participant. For each scan (premeal, satiated, and baseline), we created a first-level model that included 2 contrasts: 1) a contrast that modeled the interaction between the dlPFC time series and the BOLD response to all food stimuli (taste, sight, and combination thereof for chocolate and moldy strawberry) and 2) a contrast modeling the interaction between the dlPFC time series and the BOLD response to the control

stimuli (taste rinse, gray image, and the combination). The first eigen-time series for each participant's white matter and cerebrospinal fluid were also extracted and entered as regressors of no interest, along with motion correction variables. These were orthogonalized to the other variables. Second-level analyses were used to subtract baseline scans. A third-level analysis was then applied to examine the effect of condition (premeal vs. satiated) on the interactions. A mask of the vmPFC (10-mm radius, centered on the coordinates of the effect of condition) was applied as prethreshold masking to restrict analyses to this region. To control the FWE rate, AlphaSim was used. On the basis of our data, with a voxel-wise threshold of $P < 0.001$ (z score > 3.1), only clusters with > 13 contiguous voxels were significant with an FWE-corrected $P < 0.05$.

Behavioral analysis

Within-subject ANOVA was used to analyze data. Appetite VASs taken before and after lunch and break (for those who were satiated and premeal, respectively) were averaged over time for hunger, desire to eat, and fullness. Because all 3 items significantly correlated with each other (Pearson correlations, all $P < 0.01$), these VAS items were averaged into the composite measure "appetite" for analysis. Scanner task ratings were averaged across the scanning session and, along with region of interest data, were analyzed by condition (premeal vs. satiated), stimuli (chocolate vs. strawberry), and modality (taste, picture, and taste and picture). Only main effects of condition and interactions with condition were followed up with planned comparisons. All t tests used a Bonferroni correction for multiple comparisons. Violations of sphericity were addressed by using the Greenhouse-Geisser correction.

RESULTS

Participants

All participant scores for the Three-Factor Eating Questionnaire, STAI, Fawcett-Clark Pleasure Scale, Snaith-Hamilton Pleasure Scale, BDI, and Eating Attitudes Test questionnaires were within the normal range, and liking and craving ratings of chocolate were sufficient to ensure that chocolate would be a robust reward stimuli (see **Table 1**). In addition, there was no significant difference in the mean (\pm SE) food intake of

TABLE 2
Pleasantness, intensity, and wanting ratings for chocolate and strawberry stimuli split by modality and condition¹

	Pleasantness ²⁻⁵		Wanting ²⁻⁵		Intensity ³	
	Premeal	Satiated	Premeal	Satiated	Premeal	Satiated
Chocolate taste	1.1 ± 0.1	0.7 ± 0.1 ⁶	1.1 ± 0.1	0.5 ± 0.1 ⁶	1.5 ± 0.2	1.4 ± 0.2
Chocolate picture	1.0 ± 0.1	0.7 ± 0.1 ⁶	1.2 ± 0.1	0.5 ± 0.1 ⁶	1.2 ± 0.1	0.9 ± 0.1
Chocolate taste and picture	1.3 ± 0.1	0.9 ± 0.1 ⁶	1.3 ± 0.1	0.7 ± 0.1 ⁶	1.8 ± 0.2	1.6 ± 0.2
Strawberry taste	-1.2 ± 0.1	-1.0 ± 0.1	-1.2 ± 0.1	-1.2 ± 0.1	1.8 ± 0.2	1.6 ± 0.2
Strawberry picture	-1.0 ± 0.1	-1.0 ± 0.1	-1.3 ± 0.1	-1.2 ± 0.1	1.2 ± 0.1	1.1 ± 0.1
Strawberry taste and picture	-1.3 ± 0.1	-1.2 ± 0.1	-1.4 ± 0.1	-1.4 ± 0.1	2.0 ± 0.2	2.0 ± 0.2

¹Values are means ± SEMs; $n = 16$. Data were analyzed by using within-subject ANOVAs and paired t tests.

²⁻⁵Significant main effects: ²condition, ³stimulus, ⁴interaction between condition and stimulus, ⁵interaction between stimulus and modality.

⁶Selective effects of condition for chocolate but not for strawberry.

TABLE 3

Local maxima of key appetitive and reward areas showing a main effect of condition (satiation vs. premeal state) averaged over stimuli and modality¹

Brain region (hemisphere)	MNI coordinates			Estimated Brodmann area	z Score
	X	Y	Z		
Reduced activation when satiated (vs. premeal state)					
Ventrolateral PFC (R)	50	42	-12	47	4.0
Ventromedial PFC (L)	-12	52	-22	11	4.0
Nucleus accumbens (L)	-8	4	-12	—	3.5
Orbitofrontal cortex (R)	46	32	-12	47	3.5
Anterior medial PFC (R)	8	68	12	10	3.4
Hypothalamus (L) ²	-8	-6	-18	—	3.4
Insula (L)	-42	2	-12	48	3.1
Ventrolateral PFC (L)	-44	48	-18	47	3.1
Increased activation when satiated (vs. premeal state)					
Dorsolateral PFC (R)	46	46	16	46	4.8
Anterior lateral PFC (L)	-44	56	0	46	4.3
Parahippocampal G (L)	-16	-30	-10	30	4.3
Dorsolateral PFC (L)	-42	50	16	46	4.1
Insula (R)	38	-6	14	48	4.0
Parahippocampal G (R)	20	-18	-24	35	3.7
Anterior medial PFC	0	56	0	—	3.4

¹Values were FWE cluster corrected (voxel $P < 0.001$; cluster > 24 contiguous voxels – $P < 0.05$); $n = 16$. Data were analyzed by using a repeated-measures general linear model with the Oxford Centre for Functional Magnetic Resonance Imaging of the Brain software library (www.fmrib.ox.ac.uk/fsl). FWE, family-wise error; PFC, prefrontal cortex; G, gyrus; L, left side; MNI, Montreal Neurological Institute; R, right side.

²Hypothalamus FWE cluster corrected (voxel $P < 0.0005$; cluster > 18 contiguous voxels – $P < 0.05$).

participants in the premeal condition who ate 336.9 ± 40.5 g (1004 ± 120.9 calories) and participants in the satiated condition who ate 398.1 ± 41.8 g (1186 ± 124.6 calories) ($P > 0.05$).

Appetite VASs

There were main effects of condition [$F(1, 15) = 95.91$; $P < 0.001$] and time [$F(1, 15) = 9.63$; $P < 0.01$] and an interaction between condition and time [$F(1, 15) = 59.13$; $P < 0.001$]. There were no significant differences between baseline appetite for the satiated and premeal conditions [mean \pm SE: 52.9 ± 0.6 vs. 55.9 ± 0.5 mm; $t(15) = -0.55$, $P > 0.05$], but appetite was significantly higher in the premeal than in the satiated condition [73.3 ± 0.5 vs. 11.1 ± 0.1 mm; $t(15) = 13.37$, $P < 0.001$].

Scanner task subjective ratings

Pleasantness

Mean ratings from the scan during which participants were in the premeal or satiated state were analyzed by condition, stimuli (chocolate vs. strawberry), and modality (taste, picture, and taste

and picture). There was a main effect of condition [$F(1, 15) = 8.59$, $P < 0.05$], stimulus [$F(1, 15) = 270.36$, $P < 0.001$], and interactions between condition and stimuli [$F(1, 15) = 11.34$, $P < 0.01$] and between stimulus and modality [$F(1, 20) = 5.66$, $P < 0.05$]. Chocolate stimuli were rated significantly less pleasant when satiated than in the premeal state [$t(15) = -3.79$, $P < 0.01$; see **Table 2**]. In contrast, there were no significant differences for ratings of strawberry stimuli between the premeal and satiated conditions [$t(15) = 1.63$, $P > 0.05$].

Wanting

There was a main effect of condition [$F(1, 15) = 25.97$, $P < 0.001$], stimulus [$F(1, 15) = 268.13$, $P < 0.001$], and interactions between condition and stimulus [$F(1, 15) = 26.52$, $P < 0.001$], and stimulus and modality [$F(2, 30) = 6.96$, $P < 0.01$]. Chocolate stimuli were rated significantly less wanted in the satiated condition compared with the premeal condition [$t(15) = -6.15$, $P < 0.001$; see **Table 2**]. There were no significant differences between ratings in the premeal and satiated conditions for strawberry stimuli [$t(15) = 0.39$, $P > 0.05$].

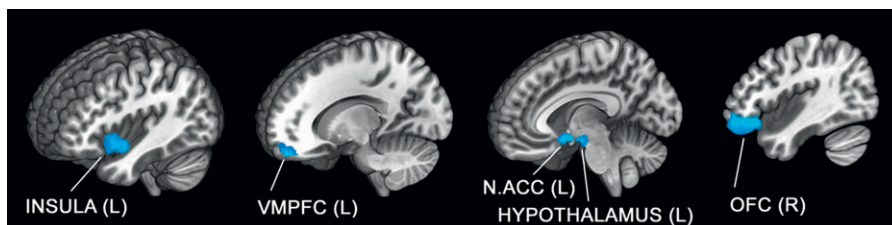


FIGURE 2 fMRI images with areas in blue showing a reduced BOLD signal when satiated (compared with premeal). L, left side; N.ACC, nucleus accumbens; OFC, orbitofrontal cortex; R, right side; VMPFC, ventromedial prefrontal cortex.

Intensity

There was a main effect of stimulus [$F(1, 15) = 15.07, P < 0.01$], in which the strawberry stimuli were rated more intense than the chocolate stimuli, and of modality [$F(1, 15) = 20.14, P < 0.001$]. There was also a trend toward an effect of condition [$F(1, 15) = 3.45, P = 0.08$], with higher intensity ratings when in the premeal state compared with the satiated condition (see Table 2).

fMRI whole-brain analysis

Baseline scans were subtracted from scans taken during the premeal and satiated states (e.g., satiated state minus baseline scan) and the contrasts [premeal (baseline subtracted) minus satiated (baseline subtracted)] were then entered into a higher-level analysis with the following factors: condition (premeal vs. satiated), stimulus (chocolate vs. strawberry), and modality (taste vs. picture vs. taste and picture). There was a main effect of condition and a main effect of modality; however, there was no main effect of stimulus and no significant interactions between any of the factors. Local maxima for the main effect of condition are reported for key appetitive/reward areas (see Table 3). Additional areas showing an effect of condition are reported in Supplemental Table 1 for reference.

Satiation attenuated BOLD activity in the vmPFC, nucleus accumbens (ventral striatum), orbitofrontal cortex, hypothalamus, and insula (see Figures 2 and 3). Activity was not attenuated in the amygdala or hippocampus at this statistical threshold. Satiation also increased activity bilaterally in the dlPFC and the insula (see Figures 4 and 5). To investigate the relation between the vmPFC and dlPFC, percentage signal change values for the local maxima were extracted and correlated for vmPFC and dlPFC while satiated. A Pearson correlation coefficient revealed a significant negative correlation ($r = -0.580, n = 16, P < 0.05$; see Figure 6). As additional checks, there were no significant differences in BOLD response between men and women, and BOLD response to the stimuli did not change over time (baseline scan day 1 compared with baseline scan day 2).

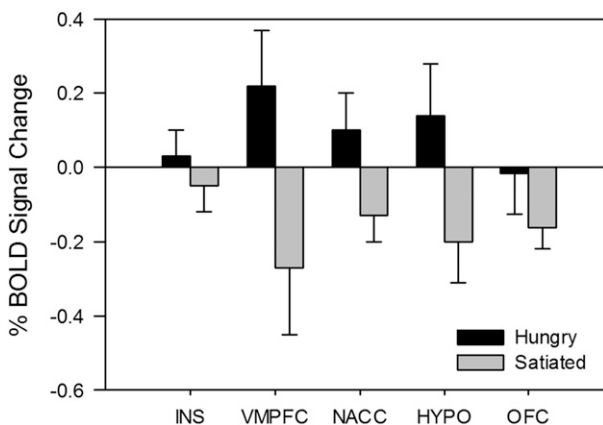


FIGURE 3 Mean (\pm SE) percentage changes in areas showing a reduced BOLD signal when satiated (compared with premeal). Clusters are FWE corrected (voxel $P < 0.001$; cluster > 24 contiguous voxels – $P < 0.05$). The hypothalamus was also FWE cluster corrected (voxel $P < 0.0005$; cluster > 18 contiguous voxels – $P < 0.05$). $n = 16$. FWE, family-wise error; HYPO, hypothalamus; INS, insula; NACC, nucleus accumbens; OFC, orbitofrontal cortex; VMPFC, ventromedial prefrontal cortex.

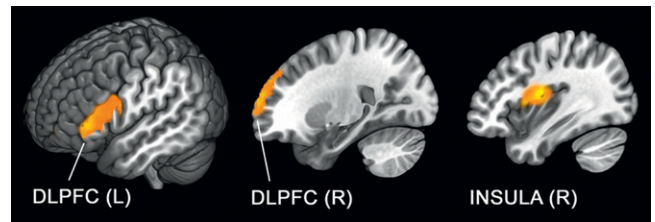


FIGURE 4 fMRI images with areas in yellow of the bilateral dlPFC and insula showing an increased BOLD signal when satiated. dlPFC, dorsolateral prefrontal cortex; L, left side; R, right side.

To further examine the relation between the dlPFC and vmPFC, a post hoc psychophysiological interaction analysis was carried out. The results confirmed that a cluster (16 contiguous voxels) in the vmPFC ($-16, 46, -18$) was more highly connected to the dlPFC when individuals were exposed to food stimuli when satiated than with a premeal state ($P < 0.001$). The same analysis for the control stimuli did not reveal a significant effect.

DISCUSSION

We used a novel fMRI paradigm to assess the effects of natural satiation on brain responses to aversive and rewarding food-related stimuli (tastes and pictures). Satiation in healthy volunteers reduced BOLD activity to all stimuli in the hypothalamus and across key reward areas. Activity in the dlPFC was increased as result of eating to satiety. Furthermore, activity in the vmPFC was negatively correlated with activity in the dlPFC and connectivity between these areas was increased in the satiated state. Importantly, this pattern of results was observed during normal eating patterns, and therefore the profile of BOLD activity is likely to reflect brain patterns associated with natural satiation. Therefore, we provide new evidence that natural satiation reduces activity in reward-related brain regions and enhances connectivity between prefrontal areas associated with higher cognitive functioning and decision making.

Multiple reward areas were attenuated by satiation, and the model appears to be sensitive to changes across the striatal-cortical pathway. Attenuation of the nucleus accumbens, vmPFC, and orbitofrontal cortex is consistent with satiety-induced decreases in reward (7, 15, 32). This is a significant finding, because previous paradigms that involve sensory-specific satiety typically report more limited profiles of attenuation [e.g., confined to the orbitofrontal cortex (15) or striatum (16)]. We also observed that natural satiation increased dlPFC BOLD activity after a meal. Interestingly, obese patients with Prader-Willi syndrome show hypoactivity in the dlPFC after a meal, which has been suggested to be related to the satiety deficit associated with this syndrome (33). It was also suggested that increased dlPFC activity reflects increased cognitive/inhibitory control (34), and Hare et al. (11) showed that the dlPFC modulates vmPFC reward responses to food, suggesting that enhancement of inhibitory control may be a mechanism of limiting further food intake by blunting reward. Our finding that there was increased connectivity between the dlPFC and vmPFC when individuals were exposed to food stimuli when satiated, and that this effect was specific to exposure to food stimuli, is consistent with the suggestion that “top down” mechanisms may contribute to reduced

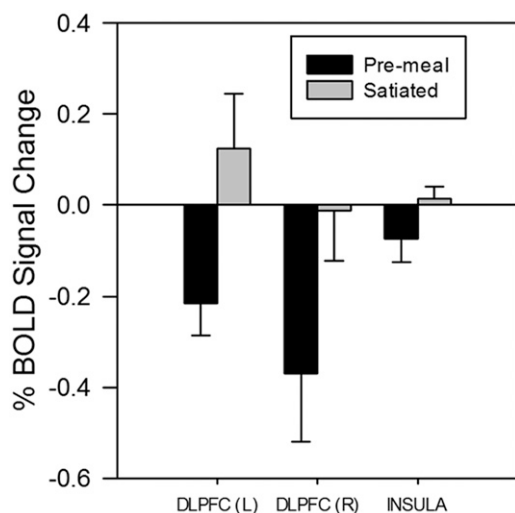


FIGURE 5 Mean (\pm SE) percentage signal changes in the left and right DLPFC and insula showing an increased BOLD signal when satiated. Clusters are family-wise error corrected (voxel $P < 0.001$; cluster > 24 contiguous voxels – $P < 0.05$). $n = 16$. DLPFC, dorsolateral prefrontal cortex; L, left side; R, right side.

motivation to eat after food consumption. However, because the dlPFC is likely to be important for a range of higher cognitive functions, such as memory (35), the specific suggestion that activity in this area reflects enhanced inhibitory control of behavior in a satiated state requires further testing, especially because we did not measure inhibitory control in this study.

For the first time, a satiation-induced decrease in hypothalamic activity was identified in response to food taste and food pictures that was not confounded by sensory-specific satiety or overeating (16, 36). It was previously reported that the administration of the anorectic drug sibutramine reduces hypothalamic responses to pictures of high-calorie foods (7). However, these authors did not observe a reduction in hypothalamic activity after participants

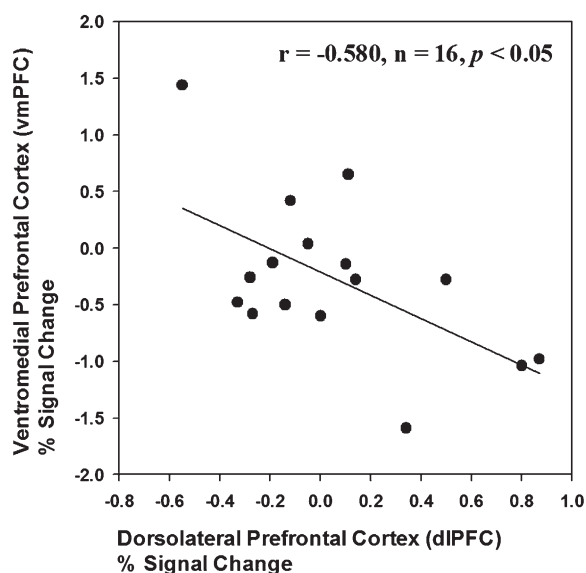


FIGURE 6 Graph displaying a negative correlation between percentage change values in BOLD signal in the dorsolateral prefrontal cortex and the ventromedial prefrontal cortex while satiated.

consumed a standard breakfast after an overnight fast. It is possible that the effects of satiety on hypothalamic response are dependent on the baseline level of hunger and whether participants are allowed to eat to fullness. Insula activity showed a more complex pattern of response, with increased superior activity and decreased inferior activity when satiated. Increased activation of the superior insula may represent an aversion to further food ingestion associated with satiation (37, 38), whereas decreased inferior insula activity may relate to bodily sensations of fullness (39).

Participants showed a decrease in wanting and pleasantness of the rewarding chocolate stimuli but not the aversive strawberry stimuli when satiated. This finding is consistent with the idea that eating-related declines in rated food pleasantness are more pronounced for hedonically positive than for aversive stimuli (40). The hedonic valence of aversive tastes may be more resistant to change because the avoidance of tastes such as sour and bitter, which normally signal the presence of toxins in food, has adaptive value (41). Although this interaction was not observed with the BOLD signal, there are several reasons why this might be the case. For instance, Berridge (42) argued that subjective ratings of reward can be distorted by conscious awareness of stimuli (i.e., cognitive processes). Hence, it is possible that the subjective ratings are not a true reflection of underlying brain activity. Alternatively, it might be that the brain regions in which satiation interacts with stimulus type (chocolate vs. strawberry) are difficult to image. For instance, the brainstem in decerebrate rats is responsive to aversive and rewarding stimuli (43) but is very difficult to image accurately in humans (44). Intensity ratings were also made by participants, and the lack of significant reductions when satiated supports evidence that the intensity of the stimulus, and potentially the neural circuitry underlying it, is dissociable from rated pleasantness and wanting (45).

In conclusion, the effect of natural satiation on brain responses to food stimuli was profiled for the first time by using a novel fMRI paradigm. Satiation decreased BOLD activity to food-related stimuli in the hypothalamus and reward-related areas, increased activity in the dlPFC, and enhanced connectivity between the dlPFC and the vmPFC. These data suggest that natural satiation is associated with a distributed pattern of neural activity suggestive of metabolic influences on both reward-related circuitry and areas involved in higher cognitive functions and decision making.

The authors' responsibilities were as follows—JMT, SH, CTD, CJH, and CM: designed the research; JMT: conducted the research; JMT, SH, and PCH: analyzed the data; JMT and SH: wrote the manuscript; and JMT, SH, and CTD: had primary responsibility for the final content. All of the authors read and approved the final manuscript. CTD is an employee and shareholder of P1vital Ltd., CJH and SH are members of P1vital's advisory panel, and JMT is funded by the Steve Cooper P1vital-BBSRC PhD Studentship. CM and PCH reported no conflicts of interest.

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