

The effects of high-intensity exercise on neural responses to images of food^{1–3}

Daniel R Crabtree, Edward S Chambers, Robert M Hardwick, and Andrew K Blannin

ABSTRACT

Background: Acute bouts of high-intensity exercise modulate peripheral appetite regulating hormones to transiently suppress hunger. However, the effects of physical activity on central appetite regulation have yet to be fully investigated.

Objective: We used functional magnetic resonance imaging (fMRI) to compare neural responses to visual food stimuli after intense exercise and rest.

Design: Fifteen lean healthy men (mean \pm SD age: 22.5 \pm 3.1 y; mean \pm SD body mass index: 24.2 \pm 2.4 kg/m²) completed two 60-min trials—exercise (EX; running at \sim 70% maximum aerobic capacity) and a resting control trial (REST)—in a counterbalanced order. After each trial, an fMRI assessment was completed in which images of high- and low-calorie foods were viewed.

Results: EX significantly suppressed subjective appetite responses while increasing thirst and core-body temperature. Furthermore, EX significantly suppressed ghrelin concentrations and significantly enhanced peptide YY release. Neural responses to images of high-calorie foods significantly increased dorsolateral prefrontal cortex activation and suppressed orbitofrontal cortex (OFC) and hippocampus activation after EX compared with REST. After EX, low-calorie food images increased insula and putamen activation and reduced OFC activation compared with REST. Furthermore, left pallidum activity was significantly elevated after EX when low-calorie images were viewed and was suppressed when high-calorie images were viewed, and these responses correlated significantly with thirst.

Conclusions: Exercise increases neural responses in reward-related regions of the brain in response to images of low-calorie foods and suppresses activation during the viewing of high-calorie foods. These central responses are associated with exercise-induced changes in peripheral signals related to appetite-regulation and hydration status. This trial was registered at www.clinicaltrials.gov as NCT01926431. *Am J Clin Nutr* doi: 10.3945/ajcn.113.071381.

INTRODUCTION

Appetite and energy intake are regulated through complex interactions between peripheral gut hormones and central receptors in the brain, which control homeostatic and nonhomeostatic feeding (1). Nonhomeostatic feeding behavior can be triggered by the hedonic nature of foods and cause overconsumption (2). This type of feeding behavior is mediated by neurons located within the orbitofrontal cortex (OFC)⁴, the insula cortex, the hippocampus, and the striatum, all of which form part of the mesolimbic reward system (3, 4). Peripheral appetite-regulating hormones, which

project signals regarding energy status to the brain, have been shown to interact with the central reward system (5, 6).

The circulation of appetite-regulating gut hormones and hunger is influenced by many factors, including physical activity (7). Acute bouts of low- to moderate-intensity exercise [40–60% maximum aerobic capacity ($\dot{V}O_{2max}$)] do not appear to greatly affect appetite (8); however, high-intensity exercise ($>70\%$ $\dot{V}O_{2max}$) has been shown to consistently suppress hunger for up to 2 h (9–11). Researchers suggest that the anorexigenic hormone peptide YY (PYY) and the orexigenic hormone ghrelin contribute toward exercise-induced hunger suppression during and after acute bouts of intense physical activity (9, 11, 12).

It is clear that exercise affects peripheral appetite regulation; however, very little is known about the effect of exercise on central appetite regulation. Recently, Cornier et al (13) examined the effect of exercise training, with and without an acute bout of moderate-intensity exercise posttraining, on neural responses to hedonic food cues in overweight participants using fMRI techniques. The authors observed that exercise training reduced neural activation when viewing pictures of food, particularly within the insula cortex. However, Cornier et al (13) reported that an acute bout of moderate-intensity exercise performed after chronic exercise training did not influence neural responses to food cues. One study has shown though that an acute bout of intense exercise does suppress neural responses to food cues within regions of the brain associated with food reward and motivation to eat, including the

¹ From the University of Aberdeen, Rowett Research Institute, Aberdeen, United Kingdom (DRC); the Department of Investigative Medicine, Imperial College London, London, United Kingdom (ESC); the Human Brain Physiology and Stimulation Laboratory, Johns Hopkins University, Baltimore, MD (RMH); and the School of Sport and Exercise Sciences, University of Birmingham, Birmingham, United Kingdom (AKB).

² All fMRI scanning costs were covered by the Birmingham University Imaging Centre pilot scanning time scheme. Pilot scanning time was granted by Stuart Derbyshire (University of Birmingham, School of Psychology).

³ Address correspondence and reprint requests to DR Crabtree, Room 3, RRS Building, Rowett Institute of Nutrition and Health, University of Aberdeen, Greenburn Road, Bucksburn, Aberdeen AB25 9SB, United Kingdom. E-mail: d.r.crabtree@abdn.ac.uk.

⁴ Abbreviations used: DLPFC, dorsolateral prefrontal cortex; EX, exercise trial; FMRI, fMRI of the brain; HC, high-calorie food; LC, low-calorie food; OFC, orbitofrontal cortex; PANAS, Positive Affect Negative Affect Scale; PYY, peptide YY; REST, resting control trial; T_{re} , rectal temperature; VAS, visual analog scale; $\dot{V}O_{2max}$, maximum aerobic capacity.

Received July 18, 2013. Accepted for publication November 12, 2013.

doi: 10.3945/ajcn.113.071381.

insula cortex and the OFC (14). However, the authors did not examine how these effects on central reward regions are related to exercise-induced changes in peripheral appetite signals. The primary aim of this study was therefore to determine the effects of an acute bout of high-intensity exercise on neural responses to viewing pictures of high-calorie food (HC) and low-calorie food (LC) and to investigate for the first time how any alterations in central responses are associated with ghrelin and PYY release. Previous neuroimaging studies have shown that pictures of HC provoke greater activation within reward regions of the brain (15, 16) and that this response is positively correlated with subjective appetite responses (15). Consequently, because of the known effects of high-intensity exercise on appetite regulatory hormones and subjective appetite ratings, it was hypothesized that the activation of reward-related brain regions to HC images would be suppressed after intense physical activity.

SUBJECTS AND METHODS

Participants

Sixteen participants originally volunteered to take part in the study; however, one participant withdrew after their first trial and was therefore excluded from the data analysis. The study received ethical approval from the Science, Technology, Engineering and Mathematics Ethical Review Committee, University of Birmingham, and the Birmingham University Imaging Centre Ethics Committee. All participants provided written informed consent and completed a general health questionnaire before their participation in the study. All participants were nonsmokers, were free from cardiovascular and metabolic disorders, were not taking any prescription medication, and regularly participated in moderate to vigorous physical activity (>2 h/wk). The $\dot{V}O_2\text{max}$ of the participants (range: 48.6–60.4 mL · kg⁻¹ · min⁻¹; mean ± SD: 54.5 ± 5.9 mL · kg⁻¹ · min⁻¹; *n* = 15) ranged from the 80th to the 99th percentile (17). The participants' characteristics are detailed in **Table 1**.

Exhaustive incremental treadmill test

Before the 2 experimental trials, each participant visited the laboratory for a preliminary session during which anthropometric measurements (height, weight, and BMI) were obtained, and an exhaustive incremental exercise test was performed. Participants were requested not to perform any physical activity 24 h before the exhaustive treadmill test. On arrival at the laboratory, height was measured using a stadiometer (Seca LTD) to the nearest 0.1 cm, and weight was measured with electronic scales (Ohaus) to the nearest 0.01 kg. An exhaustive incremental treadmill test to determine maximum aerobic capacity was then performed. This

test consisted of 5-min stages and began at 10 km/h with the treadmill speed progressively increased at each stage by 2 km/h until volitional exhaustion. Expired air samples were collected into Douglas bags (Cranlea) during the final minute of each stage for the determination of oxygen consumption and carbon dioxide production. From this test the treadmill speed needed to elicit 70% $\dot{V}O_2\text{max}$ for each subject was calculated.

Experimental trials

Exercise trials

For the exercise trials (EX), participants arrived at the Birmingham University Imaging Centre in a fasted state at 0745. On arrival the participants completed the Positive Affect Negative Affect Scale (PANAS) (18). PANAS was used to ensure that the participants' mood was similar for both trials, because mood has been shown to influence feeding behavior (19). Blood samples were taken through an intravenous cannula (BD Venflon), which was inserted into an antecubital vein. During the trial the cannula was kept patent with 2-mL flushes of 0.9% NaCl_(aq) isotonic saline solution (Baxter Health Care) after each blood-letting. Venous blood samples were drawn from the intravenous line at the beginning of the experimental trials (−10 and 0 min), at the end of EX (60 min), at the end of recovery (70 min), and immediately after the fMRI assessment (100 min). Core body temperature was monitored by using a rectal thermocouple probe (YSI 400 Series; Harvard Apparatus) inserted 12 cm beyond the external anal sphincter, which participants applied in private. The rectal thermocouple probe was attached to a data receiver (Squirrel 2020 Series; Grant), which was programmed to record mean rectal temperature (T_{re}) every minute. Heart rate was monitored during exercise by using short-range telemetry. Participants completed a 60-min run at a speed predicted to elicit 70% of their $\dot{V}O_2\text{max}$. If the participants could not maintain the required speed during the run, the speed was reduced slightly (3 participants had their speed reduced). The gradient of the treadmill remained at 0% throughout the run. After the completion of the run, the participants rested for 10 min to allow cerebral blood flow to return to resting values (20), during which time they completed the PANAS and removed the rectal thermocouple and the heart rate monitor. Participants had ad libitum access to water during the exercise and recovery periods. fMRI scans were conducted after the participants rested. After the fMRI scans were conducted, the participants completed 2 postscan tasks. The participants were shown the images during the fMRI scan, intermixed with 10 novel images, and were then asked to identify which images were shown to them during their fMRI scan. This task was performed to ensure that the participants were paying attention during their fMRI scan (6).

Control trials

The resting control trial (REST) was identical to EX; however, the participants rested for 60 min rather than exercised. Participants were asked to wear the same clothing for both trials.

Visual stimuli and experimental procedure

During each fMRI scan, food and nonfood pictures were presented to the participants on a screen in the scanner and were viewed through a mirror attached to a head coil (SENSE Head

TABLE 1
Characteristics of the participants[†]

Characteristic	All participants
Age (y)	22.5 ± 3.1 (19.4–25.6)
Height (m)	1.80 ± 0.06 (1.74–1.86)
Weight (kg)	78.7 ± 11.4 (67.3–90.1)
BMI (kg/m ²)	24.2 ± 2.4 (21.8–26.6)

[†] All values are means ± SDs; ranges in parentheses. *n* = 15.

coil 8 elements; Phillips Electronics). Participants were instructed to look carefully at each picture during the scanning. In total, 24 food and 24 nonfood visually matched (in terms of shape, complexity, brightness and color) control pictures were presented. Most of these images had previously been used in an investigation comparing the brain responses to food cues in patients with diabetes and healthy controls (21). The food pictures consisted of HC and LC. The caloric density and macronutrient profile of the HC and LC images are presented elsewhere (*see* Supplemental Tables 1 and 2 under “Supplemental data” in the online issue).

During scanning, the food and nonfood stimuli were presented in a random order by using an event-related design. Each stimulus was presented for 2 s followed by an interstimulus interval of 8 s, during which time a central fixation cross was presented. Each picture was presented twice; therefore, the total time of each scanning assessment was 960 s. The total time taken to complete a scan, including the time required to prepare the participant, was ~30 min.

fMRI data acquisition

Neuroimaging was conducted on a 3T Philips Achieva whole-body scanner (Phillips Electronics) equipped with an 8-channel SENSE head coil. A total of 480 functional whole-brain T2*-weighted images were acquired from each participant by using a 2-dimensional single-shot echo planar imaging sequence (34 axial slices, whole brain coverage, echo time = 35 ms, repetition time = 2 s, flip angle = 65°, field of view = 240 mm × 102 mm × 240 mm, 3 mm × 3 mm × 3 mm resolution). Before the fMRI assessment during rest, a high-resolution T1-weighted structural image (1 mm × 1 mm × 1 mm resolution) was also acquired for coregistration and display of the functional data. At the beginning of each functional scan, the scanning time and stimulus presentation were synchronized by a trigger signal transmitted from the fMRI scanner.

fMRI data analysis

fMRI data analysis was conducted by using FSL software library version 4.1 written by the Oxford Centre for Functional MRI of the Brain [FMRIB (22, 23)]. Motion artifacts were corrected before analysis by using FMRIB’s Linear Image Registration Tool [MCFLIRT (24)]. Nonbrain structures were removed from each participant’s structural image by using the FSL Brain Extraction Tool (25). Functional images were spatially smoothed by using a 9-mm Gaussian kernel with a high-pass filter of sigma = 20 s. A statistical parametric map was created for each participant by using a general linear model to calculate the contrast of Food > Control, HC > Control, LC > Control, HC > LC, and LC > HC at each voxel for EX and REST. All statistical parametric maps were registered to structural images and to the Montreal Neurological Institute 152 standard image using FMRIB’s Linear Image Registration Tool.

The effect of condition (EX > REST and EX < REST) on each contrast was determined by using higher level (mixed effects: FLAME 1+2) paired group analyses. Clusters of activation were formed by thresholding the z statistic image for each contrast at $z > 3.1$ (equivalent to an uncorrected 1-tailed P of 0.001). To minimize random effects, only clusters with ≥ 20 continuous voxels were retained. The criteria selected for identifying

significant clusters of activation were similar to those used by previous studies that have used fMRI techniques to examine neural responses to visual stimuli (6, 26, 27). MRIcro was used to display higher level statistical maps on a standard brain template, and the AAL atlas (28) and Talairach atlas (29) were used to identify the anatomic location of clusters of significant activation. All clusters of activation from each contrast were reported; however, only activation within the OFC, anterior insula, dorsolateral prefrontal cortex (DLPFC), hippocampus, amygdala, hypothalamus, and striatum will be discussed. These brain regions were previously shown to respond to visual food stimuli (6, 15, 26, 27, 30). In each cluster of activation, the peak voxel from the cluster was identified and the percentage change in BOLD signal extracted by using the FEATquery tool. The BOLD signal was then statistically compared in the EX and REST trials by using a paired Student’s t test performed in SPSS (SPSS Inc).

To look for nonspecific changes in fMRI activation between the EX and REST trials, BOLD signal change within the occipital lobe was compared from viewing just the control images. A mask of the occipital lobe was created from the MNI structural atlas (31), and the percentage change in BOLD signal in each trial was extracted by using the FEATquery tool and statistically compared with a paired Student’s t test performed in SPSS.

Blood sampling and analysis

For the measurement of plasma total and acylated ghrelin and total PYY (PYY₁₋₃₆ and PYY₃₋₃₆) concentrations, 3 mL whole blood was immediately added to a prechilled EDTA Vacutainer (BD Vacutainers), which had been pretreated with 0.06 mL of the serine protease inhibitor 4-(2-aminoethyl) benzenesulfonyl-fluoride hydrochloride (Alexis Biochemicals). After gentle inversion, the Vacutainers were immediately spun at 3000 × g for 10 min in a centrifuge at 4°C. After whole blood separation, 0.6-mL aliquots of the 4-(2-aminoethyl) benzenesulfonylfluoride hydrochloride-treated plasma were added to Eppendorf tubes containing 120 μ L 1N HCl and gently inverted. Treated plasma was stored at -80°C for later analysis.

Total plasma ghrelin (acylated human ghrelin and des-acyl human ghrelin), plasma acylated ghrelin, and total plasma PYY (PYY₁₋₃₆ and PYY₃₋₃₆) concentrations were measured by using commercially available sandwich ELISA kits (Millipore Corporation). A microplate reader (ELx800; BioTek) was used to measure absorbance, and standard curves were generated by plotting the absorbance unit against the standard concentrations of each ELISA to calculate the plasma concentration of each hormone. The appropriate ranges of the total ghrelin, acylated ghrelin, and total PYY ELISAs were 100–5000 pg/mL, 25–2000 pg/mL, and 14–1800 pg/mL, respectively. The intraassay CVs were 2.42%, 2.96%, and 7.88% for total ghrelin, acylated ghrelin, and total PYY, respectively. All sample measurements were performed in duplicate.

Subjective appetite sensations

Ratings of subjective appetite sensations were measured by using 100-mm visual analog scales (VASs), with extreme statements anchored at each end (ie, not at all hungry to extremely hungry). VAS were used to assess hunger, thirst, fullness, and desire to eat, as previously validated by Flint et al (32). Participants

TABLE 2

Region of activation, coordinates of peak voxel (MNI space), and peak z scores of clusters of activation formed from each contrast¹

Contrast	Region	Hemisphere	x	y	z	z Score
Food > Control						
EX > REST	Insula	R	32	20	-8	7.4
	Middle temporal gyrus	R	68	-44	2	5.7
EX < REST	OFC	L	-8	34	-10	4.1
	Precentral gyrus	L	-40	-10	30	>10.0
	Precentral gyrus	R	52	-12	26	6.1
	Hippocampus	L	-24	-38	-2	5.9
	Posterior cingulate	R	12	-58	14	>10.0
HC > Control						
EX > REST	DLPFC	L	-46	24	36	8.5
	Angular gyrus	L	-54	-58	36	5.4
EX < REST	OFC	L	-4	58	-10	7.5
	Hippocampus	L	-32	-30	-16	>10.0
	Posterior cingulate	R	6	-54	14	4.9
LC > Control						
EX > REST	Insula	L	-34	24	8	>10.0
	Insula	R	34	20	6	>10.0
	Putamen	L	-2	16	2	>10.0
	Putamen	R	22	-14	2	>10.0
EX < REST	OFC	R	10	44	-10	>10.0
	OFC	L	-8	38	-14	>10.0
	Cingulate gyrus	R	14	6	32	6.9
	Postcentral gyrus	R	38	-16	28	7
	Subgyral	L	-26	-34	42	5.2
HC > LC						
EX < REST	Pallidum	L	-22	-4	-2	>10.0
	Subgyral	L	-42	-50	-10	8.5
	Superior occipital gyrus	R	39	-78	26	5.1
LC > HC						
EX > REST	Pallidum	L	-18	-8	-6	>10.0
	Subgyral	L	-40	-54	-8	4.8

¹ Clusters were formed by thresholding the z-stat image for each contrast at $z > 3.1$ (equivalent to a 1-tailed P value of 0.001). Only clusters with >20 continuous voxels were retained ($n = 15$). DLPFC, dorsolateral prefrontal cortex; EX, exercise trial; HC, high-calorie food; L, left; LC, low-calorie food; MNI, Montreal Neurological Institute; OFC, orbito-frontal cortex; R, right; REST, resting control trial.

were presented with the VAS at the beginning of the experimental trials, after 30 min of EX/REST, at the end of EX/REST period, and immediately after the fMRI assessment. Instructions relating to the completion of the VAS were provided at the beginning of each trial. Participants were not allowed to refer to their previous VAS ratings when completing a VAS.

Statistical analysis

The sample size was based on a power calculation with the use of data from Goldstone et al (15), which compared the neural response in brain-reward systems to HC images in the fed and fasted states. With the use of the mean (\pm SD) BOLD signal within the peak voxels of the selected brain regions, it was calculated that

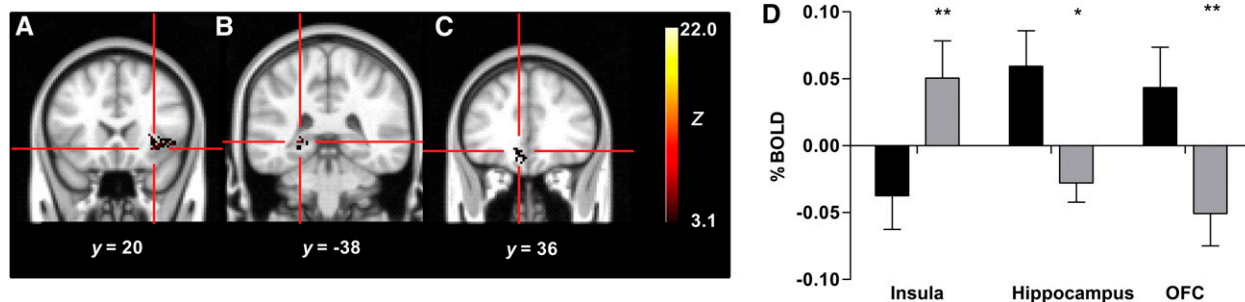


FIGURE 1. An area of the insula (A) showing significantly greater BOLD activity after EX > REST in the Food > Control contrast ($n = 15$). An area of the hippocampus (B) and OFC (C) showing significantly reduced BOLD activity after exercise compared with rest (EX < REST) in the Food > Control contrast ($n = 15$). D: Values are means \pm SEs. BOLD signal change of the peak voxel within the cluster of activation (Table 2) is highlighted in panels A, B, and C in REST (black bars) and EX (gray bars) ($n = 15$). * $P < 0.05$, ** $P < 0.01$ (paired t tests, corrected for multiple comparisons). EX, exercise trial; OFC, orbitofrontal cortex; REST, resting control trial.

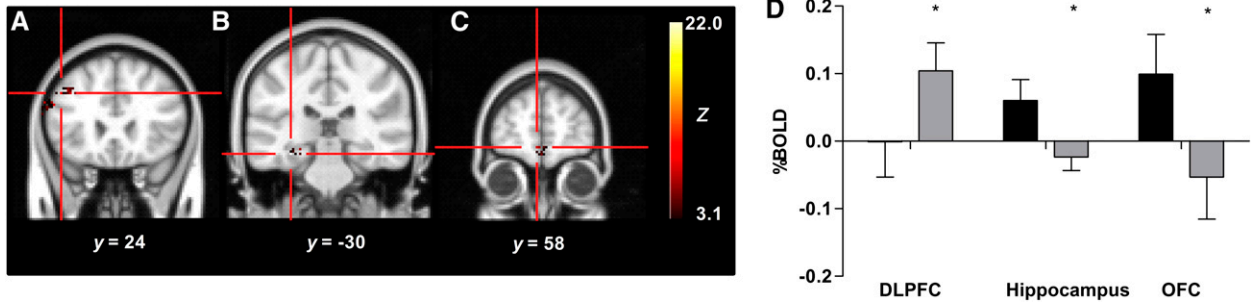


FIGURE 2. An area of the DLPFC (A) showing significantly greater BOLD activity after EX > REST in the HC > Control contrast ($n = 15$). An area of the hippocampus (B) and OFC (C) showing significantly reduced BOLD activity after EX < REST in the HC > Control contrast ($n = 15$). D: Values are means \pm SEs. BOLD signal change of the peak voxel within the cluster of activation (Table 2) is highlighted in panels A, B, and C in REST (black bars) and EX (gray bars) ($n = 15$). * $P < 0.05$ (paired t tests, corrected for multiple comparisons). DLPFC, dorsolateral prefrontal cortex; EX, exercise trial; HC, high-calorie food; OFC, orbitofrontal cortex; REST, resting control trial.

a minimum of 6 subjects would be required to observe a difference between trials with 0.80 power ($\alpha = 0.05$). Mean heart rate data were analyzed between trials by using a paired-samples t test. T_{re} data, mean positive and negative PANAS ratings, total PYY, total and acylated ghrelin, and VAS data were analyzed over time between trials by using 2-factor (trial \times time) repeated-measures ANOVA, and significant interactions were followed up with Bonferroni post hoc tests. Total PYY, total and acylated ghrelin, and VAS data were converted to AUCs by using the trapezoidal rule, and differences were analyzed by paired t tests. The relation between changes in BOLD signals of each activated brain region from each contrast and differences in gut hormones and VAS were analyzed by using Pearson correlation coefficient. The dependent factor was the individual difference in BOLD signal in the peak voxel, whereas the independent factor was the individual difference in the AUC of gut hormones and VAS ratings between the EX and REST trials. Statistical analysis was carried out with SPSS for Windows version 17.0. Data are expressed as means (\pm SDs) in the text and in tables and as means (\pm SEs) in the figures. Significant differences were accepted at $P < 0.05$, corrected for multiple comparisons.

RESULTS

Physiologic responses to EX and REST

Participants exercised at a treadmill speed of 11.7 ± 1.8 km/h during the high-intensity run. Average heart rate was significantly

higher ($P < 0.001$) during EX (159 ± 8 beats/min) than during REST (60 ± 9 beats/min). EX T_{re} was significantly higher from 10 min onward than during REST T_{re} , reaching a value of $39.3 \pm 0.86^\circ\text{C}$ at the end of EX compared with $37.0 \pm 0.48^\circ\text{C}$ after 60 min of REST ($P < 0.001$). Mean PANAS affect ratings were not significantly different between trials at any measured time point ($P > 0.05$).

fMRI data

Food > Control

During the viewing of images of food, activity in a region of the right insula was significantly greater after EX than after REST (Table 2; Figure 1A). Furthermore, EX suppressed neural activation within a region of the left hippocampus (Table 2; Figure 1B) and the left OFC (Table 2; Figure 1C) compared with REST.

HC > Control

Images of HC enhanced the BOLD response within regions of the DLPFC after EX compared with REST (Table 2; Figure 2A), whereas EX suppressed activation within a region of the left hippocampus (Table 2; Figure 2B) and left OFC (Table 2; Figure 2C) compared with REST.

LC > Control

LC images significantly increased activity in a region of the left and right insula (Table 2; Figure 3A) and in areas of the left

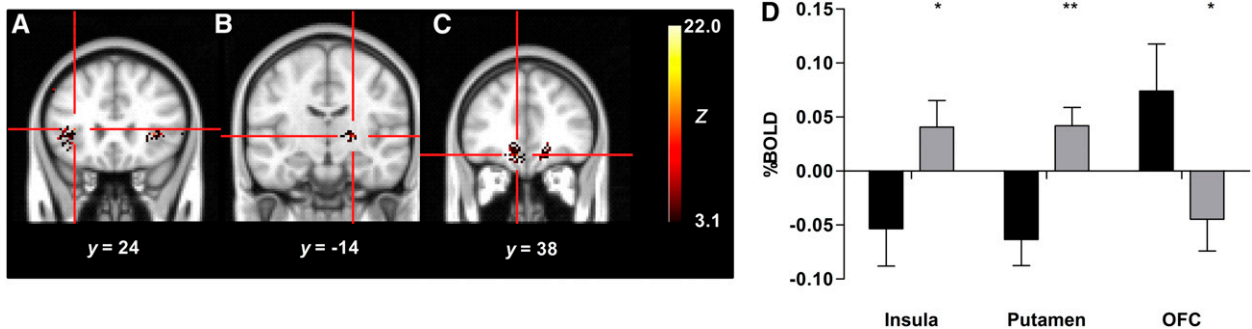


FIGURE 3. An area of the insula (A) and putamen (B) showing significantly greater BOLD activity after EX > REST in the LC > Control contrast ($n = 15$). An area of the OFC (C) showing significantly reduced BOLD activity after EX < REST in the LC > Control contrast ($n = 15$). D: Values are means \pm SEs. BOLD signal change of the peak voxel within the cluster of activation (Table 2) is highlighted in panels A, B, and C in REST (black bars) and EX (gray bars) ($n = 15$). * $P < 0.05$, ** $P < 0.01$ (paired t tests, corrected for multiple comparisons). EX, exercise trial; LC, low-calorie food; OFC, orbitofrontal cortex; REST, resting control trial.

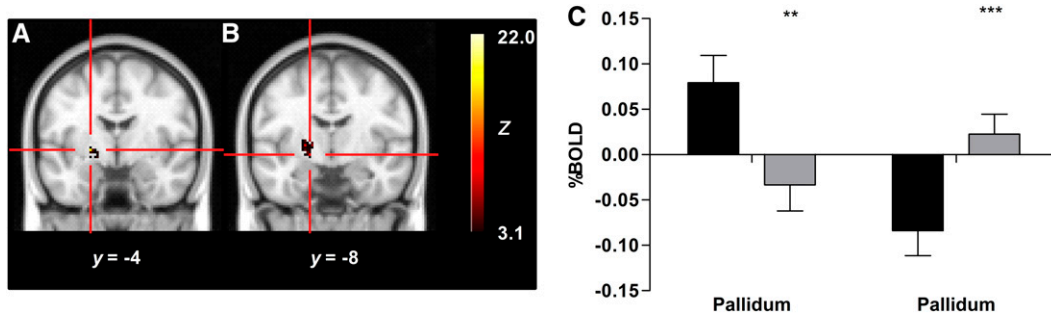


FIGURE 4. An area of the pallidum (A) showing significantly reduced BOLD activity after EX < REST in the HC > LC contrast ($n = 15$). An area of the pallidum (B) showing significantly increased BOLD activity after EX > REST in the LC > HC contrast ($n = 15$). C: Values are means \pm SEs. BOLD signal change of the peak voxel within the cluster of activation (Table 2) is highlighted in A and B in REST (black bars) and EX (gray bars) ($n = 15$). $**P < 0.01$, $***P < 0.001$ (paired t tests, corrected for multiple comparisons). $n = 15$. EX, exercise trial; HC, high-calorie food; LC, low-calorie food; REST, resting control trial.

and right putamen (Table 2; Figure 3B) after EX in comparison with REST. Furthermore, EX significantly reduced activity compared with REST in a region of the left and right OFC when images of LC were viewed (Table 2; Figure 3C).

HC > LC

After EX there was significantly less neural activation in response to HC compared with LC within a region of the left pallidum compared with REST (Table 2; Figure 4A).

LC > HC

Significantly greater neural activation within an area of the left pallidum was observed in response to LC compared with HC after EX compared with REST (Table 2; Figure 4B).

Activation of the occipital lobe from viewing the control images was not significantly different between the EX and REST trials ($P > 0.05$). In addition, no significant differences in picture recognition scores were observed between the EX ($97.2 \pm 3.8\%$) and REST ($94.5 \pm 5.1\%$) trials ($P > 0.05$).

Appetite sensations

Hunger and desire to eat scores were significantly lower during EX and immediately after EX than during REST and immediately after REST ($P < 0.01$; Figure 5, A and B). Furthermore, AUC values were significantly lower during EX than during REST for hunger (EX: $AUC_{0-100 \text{ min}} 4480 \pm 2063 \text{ mm} \times \text{min}$; REST: $6909 \pm 1751 \text{ mm} \times \text{min}$; $P < 0.001$) and for desire to eat (EX:

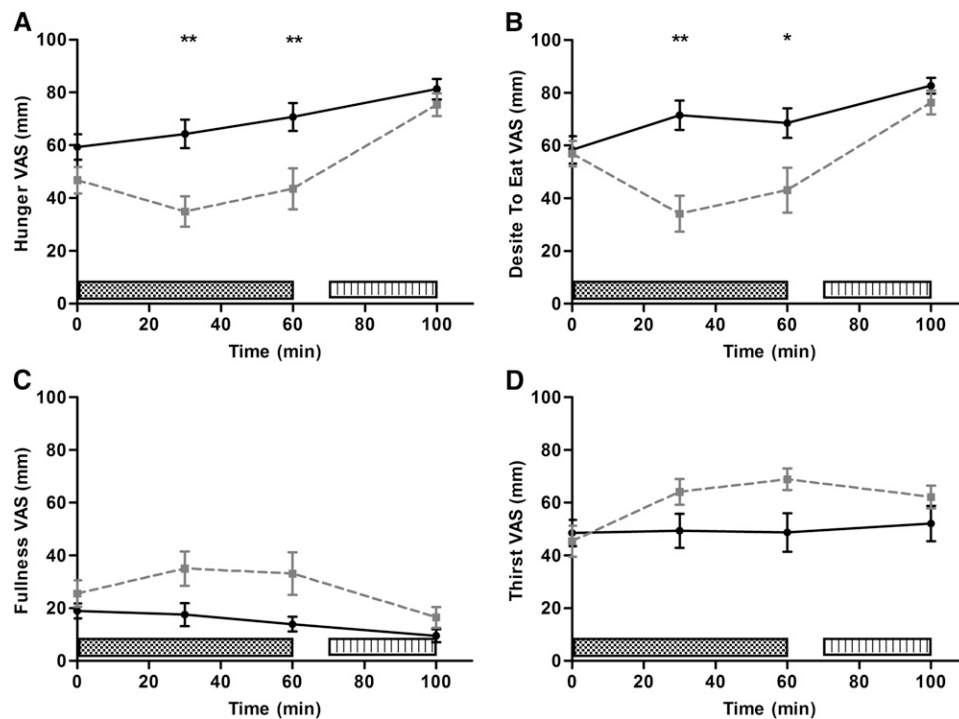


FIGURE 5. VAS scores for hunger (A), desire to eat (B), fullness (C), and thirst (D) during REST (black line) and EX (gray line) ($n = 15$). Hatched rectangles represent the treadmill run/rest; striped rectangles represent the fMRI scan. Data were analyzed by 2-factor (trial \times time) repeated-measures ANOVA. Significant interactions were followed up by Bonferroni post hoc tests. $*P < 0.05$, $**P < 0.01$. $n = 15$. EX, exercise trial; REST, resting control trial; VAS, visual analog scale.

$AUC_{0-100 \text{ min}}$ $4921 \pm 1927 \text{ mm} \times \text{min}$; REST: $7079 \pm 1638 \text{ mm} \times \text{min}$ REST; $P < 0.01$). In addition, AUC values were significantly greater during EX than during REST for fullness (EX: $AUC_{0-100 \text{ min}}$ $2913 \pm 1902 \text{ mm} \times \text{min}$; REST: $1533 \pm 1072 \text{ mm} \times \text{min}$; $P < 0.05$; Figure 5C) and thirst (EX: $AUC_{0-100 \text{ min}}$ $6257 \pm 1083 \text{ mm} \times \text{min}$; REST: $4741 \pm 2388 \text{ mm} \times \text{min}$ REST; $P < 0.05$; Figure 5D).

Appetite hormones

No significant differences in plasma total ghrelin, acylated ghrelin, or total PYY concentrations were found at baseline between trials ($P > 0.05$). Plasma total ghrelin concentrations were significantly lower immediately after EX ($P < 0.001$), before the fMRI scan ($P < 0.001$), and after the fMRI scan ($P < 0.01$) during EX than during REST (Figure 6A). Plasma acylated ghrelin concentrations were also significantly suppressed during EX compared with during REST, immediately after EX, before the fMRI scan, and after the fMRI scan (all $P < 0.001$; Figure 6B). In addition, AUC values for total ghrelin were significantly lower ($P < 0.001$) during EX ($AUC_{0-100 \text{ min}}$ $50,219 \pm 17,370 \text{ pg/mL} \times \text{min}$) than during REST ($59,819 \pm 20,838 \text{ pg/mL} \times \text{min}$) trials), as were AUC values for acylated ghrelin (EX: $AUC_{0-100 \text{ min}}$ $27,615 \pm 15,591 \text{ pg/mL} \times \text{min}$; REST: $43,358 \pm 20,446 \text{ pg/mL} \times \text{min}$ REST; $P < 0.001$). Furthermore, the percentage of total ghrelin that was acylated was significantly greater ($P < 0.001$) after EX ($65.2 \pm 11.9\%$) than after REST ($31.1 \pm 12.6\%$). Plasma total PYY concentrations were significantly greater after EX than after REST ($P < 0.01$), and PYY concentrations remained significantly elevated before the fMRI scan ($P < 0.01$) and after the scan ($P < 0.001$) during EX compared with REST (Figure 6C). Total PYY AUC values were significantly greater during EX ($AUC_{0-100 \text{ min}}$ $24,800 \pm 7141 \text{ pg/mL} \times \text{min}$) than during REST ($19,163 \pm 5938 \text{ pg/mL} \times \text{min}$; $P < 0.01$).

Correlations between gut hormones, subjective ratings of appetite, and BOLD signal

Correlation analyses were performed by using the BOLD signal of the peak voxel from every significant cluster of activation identified from each contrast (Table 2). No significant correlations were found between the difference in the peak voxel BOLD signal from any contrast and the AUC in gut hormone release. The reduced BOLD activity after EX in the left pallidum in the HC > LC contrast (Figure 7A) was negatively correlated with the AUC of subjective thirst ($r = -0.576$, $P = 0.025$), whereas the increased BOLD response in the same brain region after EX in the LC > HC contrast (Figure 7B) was positively correlated with the AUC of subjective thirst ($r = 0.625$, $P = 0.013$). No significant correlations between VAS ratings and the BOLD signal change in any other contrasts were found.

DISCUSSION

The primary aim of this study was to investigate the effects of an acute bout of intense exercise on central neural responses when images of food were viewed. Our results replicate previous studies by showing that intense physical activity significantly suppresses subjective hunger and ghrelin concentrations while increasing core-body temperature, thirst, and PYY release. The current study advances and extends previous findings by providing

the first direct evidence that these exercise-induced changes in peripheral signals related to appetite-regulation and hydration status are associated with altered neural responses within brain-reward regions to images of HC and LC.

The Food > Control contrast showed that images of food reduced neural activity within the OFC and hippocampus after EX. It was previously shown that these areas of the brain regulate the drive to eat and process the rewarding aspects of foods (33–35). The primary role of the hippocampus is memory retrieval (36); however, this area of the brain also contributes to appetite regulation (16, 35) and has been implicated in reward system interaction when pictures of HC are viewed (15). The OFC assigns reward values to stimuli, such as HC, by communicating with the amygdala and the anterior cingulate cortex (30). Furthermore,

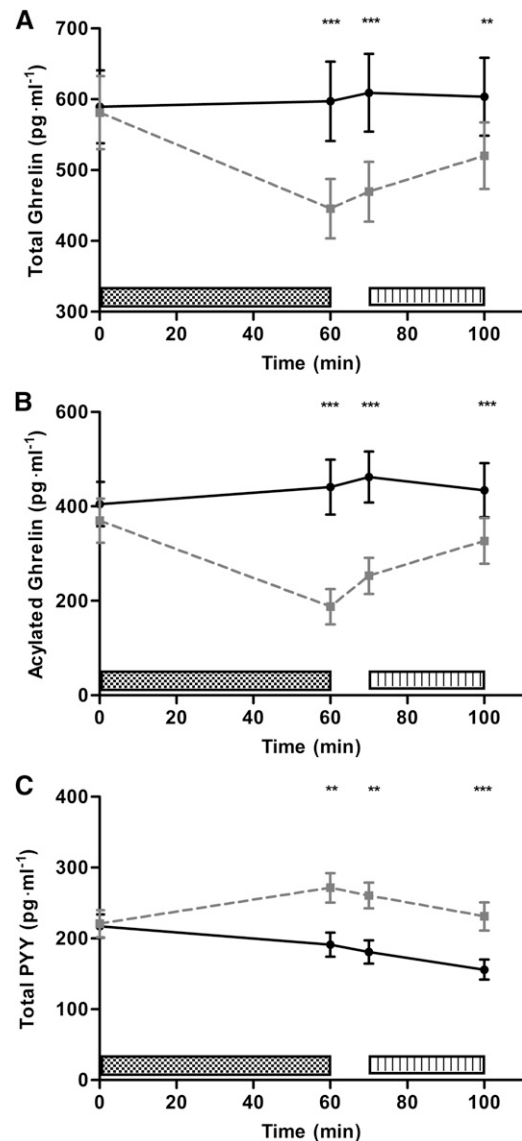


FIGURE 6. Plasma total ghrelin (A), acylated ghrelin (B), and total PYY (C) concentrations during REST (black line) and EX (gray line) ($n = 15$). Hatched rectangles represent the treadmill run/rest; striped rectangles represent the fMRI scan. Data were analyzed by 2-factor (trial \times time) repeated-measures ANOVA. Significant interactions were followed up by Bonferroni post hoc tests. * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$. $n = 15$. EX, exercise trial; PYY, peptide YY; REST, resting control trial.

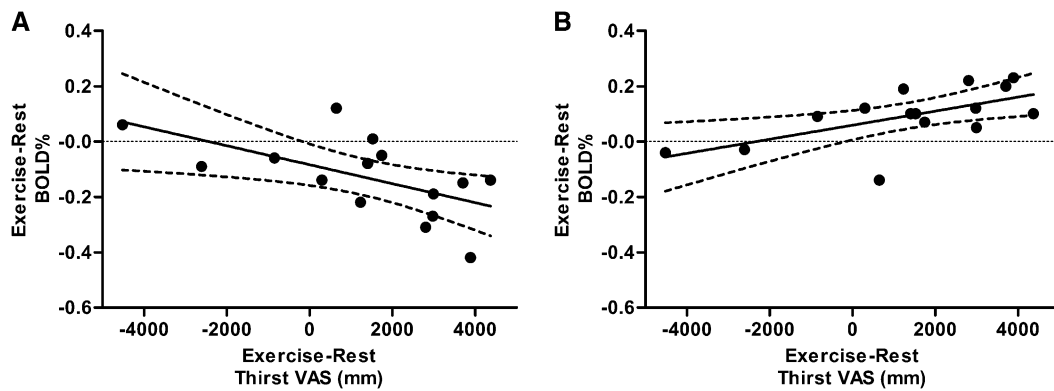


FIGURE 7. A: Reduced BOLD activity in the left pallidum after EX in the HC > LC contrast (Figure 4A) is negatively correlated ($r = -0.576$, $P = 0.025$) with the difference in $AUC_{0-100min}$ of subjective thirst ($n = 15$). B: Increased BOLD activity in the left pallidum after EX in the LC > HC contrast (Figure 4B) is positively correlated ($r = 0.625$, $P = 0.013$) with the difference in $AUC_{0-100min}$ of ratings of thirst ($n = 15$). Dashed lines represent the 95% CIs of the solid regression line. $n = 15$. EX, exercise trial; HC, high-calorie food; LC, low-calorie food; REST, resting control trial; VAS, visual analog scale.

enhanced OFC activity in obese individuals in response to images of HC has been suggested to contribute to hyperphagia (16). In the current study, activity within the OFC was significantly lower after EX in the HC > Control contrast. The reductions in OFC activity after physical activity may have been associated with the significantly increased PYY concentrations and reduced total and acylated ghrelin concentrations found during EX. Whereas we did not observe any direct correlations with the peak voxel of activation in the OFC in the Food > Control and HC > Control contrasts, previous studies have reported that exogenous administration of PYY and ghrelin modulates the fMRI response to visual food cues within the OFC and other central reward regions (6, 37). PYY was shown to reduce BOLD activation (37) and ghrelin was shown to increase BOLD activation (6) within the OFC, in keeping with the proposed contrasting physiologic effects of these gut hormones on feeding behavior (1).

The current study observed suppression in sensations of hunger and desire to eat during and immediately after EX. This finding agrees with that of several studies, which have shown a transient decline in appetite during and after high-intensity exercise (9–11)—a phenomenon termed exercise-induced anorexia (38). Therefore, one might expect to observe an increase in BOLD response within central regions that signal satiety and meal termination. After the bout of exercise, DLPFC activity was increased in the HC > Control contrast. The DLPFC was previously shown to regulate inappropriate behavior (39) and to promote satiation by transmitting inhibitory signals to appetite-stimulating areas of the brain, such as the insular cortex and the OFC (34, 35). Indeed, in the current study neural activity within the OFC and the hippocampus was suppressed in the HC > Control contrast after EX. Le et al (39, 40) showed that postprandial DLPFC activation was suppressed in overweight compared with lean subjects, which indicated that blunted DLPFC activation may be associated with overconsumption and weight gain. The current study suggests that intense physical activity may acutely increase activation in brain regions associated with inhibitory control in response to HC visual food stimuli. Previous studies have reported activation within the hypothalamus when viewing HC images (16, 37). Despite significant differences between trials in subjective appetite responses, we found no differences in activation within the hypothalamus in any statistical contrast. This may have been attributable to the

study lacking the power to observe differences within the small hypothalamic nuclei involved in appetite regulation.

A comparison of neural responses with the LC > Control contrast showed that postexercise activation within the putamen and the insula was enhanced, whereas OFC activity was suppressed. The putamen forms part of the striatal region within the basal ganglia, and putamen activation has been observed when viewing pictures of foods that are perceived as rewarding (16, 41); therefore, it has been proposed that the putamen may form part of the central network that processes the motivational value of foods (42). The insula is recognized as the primary taste cortex (30), and previous research has shown that anterior insula activity is increased in anticipation of rewarding foods (43) and when foods that are considered pleasant are consumed (34, 44). In the current study, despite the participants' ad libitum access to water, subjective thirst was significantly higher during EX. Therefore, because of the role that the left insula plays in the regulation of thirst (45), it could be speculated that the LC > Control contrast had greater activation within the insula and putamen after EX because participants perceived the LC as having a high water content (eg, fruit and salad vegetables) and therefore able to satisfy exercise-induced thirst. Furthermore, left pallidum activation after EX decreased in the HC > LC contrast and increased in the LC > HC, and this BOLD activity was significantly correlated with subjective thirst. The pallidum is reported to be the final pathway for limbic "liking" and "wanting" signals and receives projections from regions of the brain that motivate reward-related feeding (46). Previous researchers have shown that the body's drive to maintain fluid balance can be more powerful than the drive to maintain energy balance (47). In addition, research suggests that compensating for the energy expended during exercise may be delayed by slowing the feeding response; instead, priority is given to maintaining fluid balance (48). Therefore, the current study suggests that the postexercise increase in BOLD responses within brain-reward regions to LC visual food cues may have been related to exercise-induced fluid loss and the body's homeostatic preference for maintaining fluid balance ahead of energy balance. A limitation of the current study was that water intake and sweat loss were not quantified during EX. Nevertheless, previous studies suggest that participants would replace 65–75% of fluid losses with ad libitum fluid intake during exercise; thus, any differences in fluid balance between

participants after 1 h of exercise in a thermoneutral environment would be small (49). However, future research should control for food intake and sweat losses and use other techniques (hematocrit/osmolality) in combination with subjective thirst to fully investigate the effect of hydration status and fluid balance on central neural responses to visual food stimuli.

During EX, oxygen consumption could not be measured because the study was conducted within an fMRI facility and not in an exercise laboratory. However, the average heart rate during exercise was 159 ± 8 beats/min, which indicates that the run was performed at a suitably high intensity. Furthermore, changes in appetite sensations and appetite hormone concentrations were similar to those that were observed previously during and after an acute bout of treadmill running performed at 70% $\dot{V}O_2\text{max}$ (9, 10). Another limitation of the current study was that it did not directly measure energy intake at any point during the study; therefore, it is not possible to relate the exercise-induced changes in markers of appetite to actual food intake. A further limitation of the current research is that only lean physically active men were studied, because previous studies have suggested that the response of central reward regions to images of HC is greater in obese individuals than in lean controls (16, 50). Consequently, neural responses to HC stimuli after high-intensity exercise may vary between individuals with different body composition. In addition, this study only measured central appetite responses immediately after exercise. Research has shown that reductions in sensations of appetite after high-intensity exercise are short lived (51), and appetite responses may rebound over several hours after exercise (9, 10). The effect of physical activity on the central responses to food stimuli in the hours after exercise warrants further investigation.

In conclusion, the current study advances understanding of the complex links between physical activity, energy balance, and peripheral and central appetite regulation. Central reward system activation was increased in response to images of LC after high-intensity exercise, whereas responses to HC images were suppressed. These central neural responses were allied with exercise-induced changes in appetite regulatory hormones and increases in subjective thirst.

We thank Stuart Derbyshire for granting us the fMRI scanning time required to conduct this study. We also thank Nina Salman for conducting the fMRI scanning and Charnjit Sindu for providing assistance with the fMRI data analysis. Finally, we thank the volunteers for participating in this study.

The authors' responsibilities were as follows—DRC: contributed to the research design, conducted the data collection, analyzed the data, wrote the manuscript, and had primary responsibility for the final content; ESC: conceived the study, contributed to the research design, contributed to the data collection, provided essential materials, provided data analysis assistance, and wrote the manuscript; RMH: provided essential materials, contributed to the data analysis, and helped write the manuscript; and AKB: supervised the project, contributed to the research design, conducted the data collection, provided essential materials, provided data, assisted with the analysis, and wrote the manuscript. There were no conflicts of interest reported.

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