Title

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Journal

Gastroenterology, 146(5)

ISSN

0016-5085

Authors

Kilpatrick, Lisa A
Coveleskie, Kristen
Connolly, Lynn
et al.

Publication Date

2014-05-01

DOI

10.1053/j.gastro.2014.01.023

Peer reviewed

Published in final edited form as:
Gastroenterology. 2014 May ; 146(5): 1212–1221. doi:10.1053/j.gastro.2014.01.023.

Influence of Sucrose Ingestion on Brainstem and Hypothalamic Intrinsic Oscillations in Lean and Obese Women

Lisa A. Kilpatrick[1,2], **Kristen Coveleskie**[1], **Lynn Connolly**[1,2], **Jennifer S. Labus**[1,2,3,5], **Bahar Ebrat**[1,2], **Jean Stains**[1,2], **Zhiguo Jiang**[1,2], **Brandall Y. Suyenobu**[1,2], **Helen E. Raybould**[6], **Kirsten Tillisch**[1,2], and **Emeran A. Mayer**[1,2,3,4,5]

[1]Gail and Gerald Oppenheimer Family Center for Neurobiology of Stress, David Geffen School of Medicine, University of California, Los Angeles

[2]Department of Medicine, Division of Digestive Diseases, University of California, Los Angeles

[3]Ahmanson-Lovelace Brain Mapping Center, Los Angeles, California

[4]Department of Psychiatry, University of California, Los Angeles, Los Angeles, California

[5]Brain Research Institute, University of California, Los Angeles, Los Angeles, California

[6]Department of Anatomy, Physiology, and Cell Biology, School of Veterinary Medicine, University of California, Davis, Davis, California

Abstract

BACKGROUND & AIMS—The study of intrinsic fluctuations in the blood oxygen level-dependent signal of functional magnetic resonance imaging can provide insight into the effect of physiologic states on brain processes. In an effort to better understand the brain–gut communication induced by the absorption and metabolism of nutrients in healthy lean and obese individuals, we investigated whether ingestion of nutritive and non-nutritive sweetened beverages differentially engages the hypothalamus and brainstem vagal pathways in lean and obese women.

METHODS—In a 2-day, double-blind crossover study, 11 lean and 11 obese healthy women underwent functional magnetic resonance imaging scans after ingestion of 2 beverages of different sucrose content, but identical sweetness. During scans, subjects rested with eyes closed.

RESULTS—Blood oxygen level-dependent fluctuations demonstrated significantly greater power in the highest frequency band (slow-3: 0.073–0.198 Hz) after ingestion of high-sucrose compared with low-sucrose beverages in the nucleus tractus solitarius for both groups. Obese women had greater connectivity between the right lateral hypothalamus and a reward-related brain region and weaker connectivity with homeostasis and gustatory-related brain regions than lean women.

CONCLUSIONS—In a functional magnetic resonance imaging study, we observed sucrose-related changes in oscillatory dynamics of blood oxygen level-dependent fluctuations in brainstem

Address requests for reprints to: Lisa A. Kilpatrick, PhD, Oppenheimer Family Center for Neurobiology of Stress, 10833 Le Conte Avenue, CHS 42-210 MC 73781, Los Angeles, CA 90095-7378. lakilpatrick@mednet.ucla.edu; fax: (310) 825-1919.

Conflicts of interest
The authors disclose no conflicts.

and hypothalamus in lean and obese women. The observed frequency changes are consistent with a rapid vagally mediated mechanism due to nutrient absorption, rather than sweet taste receptor activation. These findings provide support for altered interaction between homeostatic and reward networks in obese individuals.

Keywords

Resting State; Obesity; Satiety; Food Intake

Increased engagement of the reward-based brain networks and reduced reliance on interoceptive input has been suggested in the pathophysiology of some forms of obesity ("food addiction").[1] Functional neuroimaging studies of obesity and food intake regulation have typically compared brain activity to visual food cues after ingestion of a meal with brain activity in a fasting state, or compared ingestion of a glucose solution with ingestion of water.[2] Although the glucose solution and water share similar oral somatosensory features and distension volume (unlike meal and fasting), the comparison of glucose and water does not allow specific statements regarding the brain's response to the absorption of nutrients in the gut. Glucose compared with water differentially activates lingual and intestinal sweet taste receptors, as well as pathways that depend on absorption and metabolism of glucose.[3–6]

In an effort to better understand brain–gut communication driven by the absorption and metabolism of nutrients in healthy and obese individuals, we used a paradigm involving ingestion of 2 beverages designed to be similar in sweetness but different in sucrose content. We compared ingestion of a high-sucrose beverage with a control condition of a low-sucrose beverage containing a non-nutritive sweetener to make the 2 drinks indistinguishable in terms of sweetness. These beverages should both activate sweet taste receptors but differ in vagal stimulation due to the absorption and metabolism of sucrose within the gut.

A novel aspect of this study concerns the type of functional neuroimaging and analysis performed. The dominant design of functional magnetic resonance imaging (fMRI) studies to date has involved an active task condition. For the study of food intake regulation, food images are often presented as an external cue to activate the brain. However, it is now appreciated that a wealth of information can be extracted from the fluctuations in the blood-oxygen level–dependent (BOLD) signal that are seen in the brain even when the subject is at rest and not actively engaged in discrete tasks, ushering new approaches to fMRI design and analysis. Although several analysis approaches have been proposed to analyze changes in intrinsic brain oscillations, one approach focuses on frequency power in specific frequency bands within the range detectable in the BOLD signal thought to represent different neuronal oscillation classes.[7,8] Although the precise relationship between oscillatory brain dynamics and behavior remains to be established, this approach has been applied to resting-state fMRI by a growing number of researchers.[9–12]

By measuring sucrose-related intrinsic BOLD activity compared with a control condition of non-nutritive sweetener ingestion, we sought to monitor brain activity related to vagal stimulation triggered by various peripheral glucose-sensing mechanisms dependent on the metabolism and absorption of sugar, and to identify obesity-related changes in brain–gut

communication. We used the novel tool of measuring oscillatory brain dynamics in multiple frequency bands to test 2 specific hypotheses, that ingestion of sucrose is associated with engagement of hypothalamus and brainstem vagal pathways, as reflected in changes in specific frequency bands; and obese and lean subjects differ in their central response to sucrose ingestion. The results confirmed both hypotheses.

Methods

Subjects

Eleven lean healthy women (body mass index 19–25) and 11 obese healthy women (body mass index 30–37) were recruited by advertisement. All subjects were right-handed. Subjects were interviewed by a nurse practitioner for absence of significant health or psychiatric conditions. Subjects diagnosed with diabetes were excluded. All subjects were regularly menstruating and were scanned 4–12 days after the first day of last menstrual period. Study protocols were performed after approval by the review committee at University of California, Los Angeles's Office of Protection for Research Subjects and informed consent was obtained from all subjects.

Behavioral Measures

All subjects completed the Hospital Anxiety and Depression Scale, a measure of current anxiety and depression symptoms validated for nonpsychiatric samples.[13] Subjects also completed visual analog scales (VAS) to rate the taste of the beverages and assess hunger (How hungry do you feel?), satisfaction (How satisfied do you feel?), and desire to eat something sweet (Would you like to eat something sweet?).[14] Subjects were asked to make a mark across a 10-cm line representing the magnitude of their response. Qualifying statements were provided on the extreme left and right side of the 10-cm line. VAS scores were quantified by measuring the distance of the mark (in cm; range, 0–10) relative to the end of the line associated with a low/poor response; a high score reflected a high/good response for all VAS items.

Procedures

Procedures are summarized in Figure 1. Subjects underwent 2 separate days of fMRI (1.5T) scanning. Subjects fasted, drinking only water, for 6 hours before the scan appointment. Scans occurred during the morning, between 9:00 AM and 11:00 AM. Ten minutes before functional scanning, subjects consumed 296 mL of a beverage consisting of a low-calorie carbohydrate drink (Diet Ocean Spray Cranberry Juice with 50 g of Truvia; <10 calories; low sucrose) or a high-calorie carbohydrate drink (Ocean Spray Cranberry Juice with 50 g of sugar; 300 calories; high sucrose). The beverages were designed to be similar in taste and sweetness. Pilot testing in 5 healthy individuals confirmed that the drinks could not be differentiated on the basis of taste. Subjects completed the VAS questionnaires immediately before beverage consumption. Subjects consumed a different beverage on each scan day and drink order was counterbalanced with subject and investigators blinded to drink order. The beverage was presented in a nondescriptive container. Subjects drank through a straw and were fitted with a nose clip during consumption to minimize the flavor of the juice. Also, 1 oz of water was used to cleanse the palate. After beverage consumption, subjects again

completed the VAS questionnaires and were encouraged to use the bathroom. Ten minutes after beverage consumption, subjects completed a 10-minute functional scan while resting with eyes closed.

fMRI Acquisition

MRI scanning was performed using a 1.5T MRI scanner (Siemens Sonata; Siemens, Erlangen, Germany). A high-resolution structural image was acquired from each subject with a magnetization-prepared rapid gradient-echo sequence, repetition time = 2200 ms, echo time = 4.38 ms, slice thickness = 1 mm, 176 slices, 256 × 256 voxel matrices, and 1^3 mm voxel size. Subjects rested with eyes closed while functional BOLD images were acquired for 10 minutes in 24-slice whole brain volumes, slice thickness = 5 mm, repetition time = 2000 ms, echo time = 45 ms, flip angle = 77 degrees. After the resting scan, subjects completed additional fMRI scans involving food-related images; these data are not included in the current report, and is reported elsewhere.[15]

fMRI Preprocessing

Using Data Processing Assistant for Resting-State fMRI[16] (http://www.restfmri.net/forum/DPARSF), which is based on SPM8 (Welcome Department of Cognitive Neurology, London, UK) and Resting-State fMRI Data Analysis Toolkit,[17] data were slice-time and motion corrected. Nuisance covariate regression was then performed to minimize physiological noise using 6 head-motion parameters, white matter signal, and cerebral spinal fluid signal. Data were spatially normalized to the Montreal Neurological Institute template using structural scans. Spatial smoothing with a 3 mm^3 Gaussian kernel occurred after calculation of frequency and functional connectivity maps (see Statistical Analyses).

Statistical Analyses

Frequency analysis—Using the framework developed by Buzsaki and colleagues,[7,8] the BOLD signal was subdivided into 3 frequency bands, referred to in the literature as slow-5 (0.01–0.027 Hz), slow-4 (0.027–0.073 Hz), and slow-3 (0.073–0.198 Hz). For convenience, we will refer to these 3 frequency bands as low frequency (LF), medium frequency (MF), and high frequency (HF), respectively. A gray-matter mask was applied to restrict analyses to gray-matter regions. Relative power within each of the 3 frequency bands was computed for each voxel in the brain using fractional amplitude of low-frequency fluctuations modified for our frequency ranges.[18] A Group (Lean; Obese) × Condition (High Sucrose; Low Sucrose) flexible factorial analysis was performed for each band in SPM8 with age and depression entered as covariates to identify sucrose-related and group-related differences in the distribution of frequency power. Region of interest analyses were performed using hypothalamus and brainstem templates produced in WFU PickAtlas.[19] A cluster-level familywise error corrected $P < .05$ was considered significant. The mean value from each significant cluster was extracted by the marsbar toolkit for SPM for further evaluation.[20] As relative frequency was measured, a post-hoc analysis of variance (ANOVA) was performed in PASW v17.0 (Chicago, IL) on regions of interest to see if changes in one band were accompanied by changes in other measured bands. All functional data were overlaid on the

Montreal Neurological Institute template available in MRIcron (http://www.cabiatl.com/mricro/mricron/index.html) for presentation purposes.

Seed-based correlation analysis—Seed-based correlation is a widely used approach in the study of resting state connectivity with advantages over independent components analysis in terms of investigating functional connectivity of specific regions of interest.[21] Functional data was bandpass filtered using LF, MF, and HF bands and band-specific fisher transformed maps of the bivariate correlation between seed (identified in the frequency analysis) time-courses and all other voxels were created using Data Processing Assistant for Resting-State fMRI. Nonrotated behavioral partial least squares analysis (PLS) was applied to the normalized correlation maps to extract group-related differences in functional connectivity of the nucleus tractus solitaries (NTS) and hypothalamus identified in the frequency analysis. PLS is a multivariate statistical technique similar to principal components analysis with the solutions restricted to the part of the covariance structure that is attributable to experimental design and is more sensitive than SPM.[22] PLS was implemented with freely available code (www.rotman-baycrest.on.ca/pls/). The PLS analysis produced 2 spatial maps, 1 associated with group differences across sucrose condition and 1 associated with group differences exaggerated during the high-sucrose condition. Voxel reliability on each of the maps was determined using bootstrap estimation (500 samples). The ratio of the observed weight to the bootstrap standard error was calculated and voxels were considered reliable if the absolute value of the bootstrap ratio exceeded 3.30 ($P < .001$).

Behavioral analyses—Behavioral analyses were performed in PASW v17.0. Group differences in Hospital Anxiety and Depression Scale anxiety and depression ratings were evaluated by independent samples *t* tests. Taste ratings were evaluated in a Group (Lean; Obese) × Condition (High Sucrose; Low Sucrose) ANOVA. Appetite ratings were evaluated in Group (Lean; Obese) × Condition (High Sucrose; Low Sucrose) × Time (Before Consumption; After Consumption) repeated measure ANOVA.

A linear mixed model approach was implemented in SAS 9.2 software (SAS Institute, Cary, NC) to estimate the correlation, adjusted for repeated measures, between appetite/taste ratings and frequency power in hypothalamus and NTS clusters identified in the frequency analysis.[23]

Results

Behavioral and Subject Data

Mean age, body mass index, and ratings are provided in Table 1. Obese subjects had significantly higher depression symptom ratings (all within the normal range of the Hospital Anxiety and Depression Scale) compared with lean subjects ($t[20] = 2.242$; $P = .036$), but did not differ in anxiety ratings ($P > .05$). Obese subjects rated the taste of both beverages significantly lower than lean subjects ($F[1] = 8.617$; $P = .005$). Beverage consumption significantly reduced hunger ratings ($F[1] = 8.681$; $P = .005$), increased satisfaction ratings ($F[1] = 52.376$; $P < .001$) and reduced desire for sweetness ($F[1] = 40.159$; $P < .001$) across all subjects, without any significant group or calorie condition differences.

Sucrose and group effects on temporal dynamics of BOLD oscillations—A change in the frequency distribution during the high-sucrose condition (300-calorie beverage) compared with the low-sucrose condition (<10-calorie beverage) in both lean and obese subjects was demonstrated in several regions of the brainstem encompassing the NTS, the periaqueductal gray, locus coeruleus, as well as other pontine and medullary regions. The NTS region demonstrated significantly greater HF power (slow-3) during the high-sucrose condition compared with the low-sucrose condition in both lean and obese subjects, and the other brainstem regions demonstrated significantly less LF power (slow-5) during the high-sucrose condition compared with the low-sucrose condition in both lean and obese subjects (Figure 2; Table 2).

An interaction was demonstrated for a small cluster in the right hypothalamus (Figure 2; Table 2). Although lean women had decreased hypothalamus LF power during the high-sucrose condition compared with the low-sucrose condition, obese women had increased hypothalamus LF power during the high-sucrose condition. However, the interaction was only a trend once multiple comparison correction was applied.

A significant interaction was demonstrated for the hypothalamus (Figure 3, Table 2). Although lean women had increased slow-5 frequency power during the low-sucrose condition compared with obese women, obese women had increased slow-5 frequency power during the high-sucrose condition compared with lean women. In addition, a significant group difference in frequency power was demonstrated for a cluster within the periaqueductal gray (Figure 3; Table 2). Lean women demonstrated significantly greater slow-5 power than obese women, regardless of drink condition. Examination of the cluster means revealed that this region is more clearly dominated by slow-3

Given their prominent role in central regulation of ingestive behavior, the NTS and hypothalamus were selected as region of interests for additional analyses. Examination of the mean power in each band for NTS and hypothalamus revealed a redistribution of frequency power for NTS. The increased HF during high sucrose compared with low sucrose was accompanied by decreased LF/MF power (Figure 3*A*), while sucrose-related changes in frequency power for the right hypothalamus were seen only in the LF band (Figure 3*B*). Sucrose and group-related differences in functional connectivity, as well as the relationship between temporal dynamics and subjective ratings, were examined for NTS and right hypothalamus. In addition, NTS and hypothalamus frequency power during the first half of the imaging session (10–15 minutes postprandially) were compared with the second half (15–20 minutes postprandially) of the imaging session to gain a slightly more fine-tuned temporal profile of oscillatory dynamics in these brain regions.

Temporal profile of NTS and hypothalamus oscillatory dynamics—Mixed model ANOVAs revealed no significant differences in NTS and hypothalamus frequency power in the first half of the imaging session compared with the second half ($P > .05$). NTS HF power was significantly increased during the high-sucrose condition compared with the low-sucrose condition in both the first and second half of the imaging session ($F[1,20] = 15.85$; $P = .001$ and $F[1,20] = 9.14$; $P = .007$; respectively). Likewise, hypothalamus LF power demonstrated significant Group × Sucrose condition interactions in both the first and second

half of the imaging session ($F[1,20] = 10.71$; $P = .004$ and $F(1, 20) = 4.55$; $P = .045$; respectively).

Relationship between NTS and hypothalamus frequency power and subjective ratings—Obese women, but not lean women, demonstrated significant negative correlations between hypothalamus LF power and taste ratings ($r = -0.44$; $P = .04$) and between NTS HF power and taste ratings ($r = -0.51$; $P = .02$). No other correlations reached significance ($P > .05$).

Functional connectivity of the NTS and hypothalamus—The functional connectivity of the right hypothalamus with posterior insula, the primary interoceptive cortex, and parietal cortex (BA43) was more positive in lean women compared with obese across both sucrose conditions (Table 3). In contrast, the functional connectivity of the hypothalamus with the putamen, a reward-related regions, as well as the cerebellum, was more positive in obese women compared with lean. During high sucrose, the differences between the groups increased with greater, more positive hypothalamus functional connectivity with posterior insula, parietal cortices (BA43, BA 40, BA41, BA7) and posterior cingulate cortex (PCC) in lean women and greater hypothalamus connectivity with putamen, BA10, and cerebellum in obese women. Functional connectivity of the NTS also demonstrated group differences with more positive connectivity with the PCC, BA22, and cerebellum in lean women compared with obese across sucrose conditions (Table 3). The differences in connectivity of the NTS with PCC did not become exaggerated during high-sucrose ingestion; however, group differences in NTS connectivity with BA7 and BA8 emerged with lean women demonstrating greater, more positive connectivity compared with obese women.

Discussion

The current study examined differences in intrinsic brain activity related to sucrose ingestion occurring 10–20 minutes after consumption of 2 test drinks. Ingestion of a high-sucrose, compared with a low-sucrose drink of similar taste and sweetness was associated with differences in the oscillatory dynamics in brainstem regions, including an area encompassing the NTS and to some extent in the right hypothalamus. The NTS is the main recipient of vagal afferents from the gut and forms the main neural conduit through which visceral afferent input affects homeostasis.[24] This effect was seen in the first 5 minutes of the imaging session (10–15 minutes after beverage consumption) as well as in the second 5 minutes of the imaging session (15–20 minutes after beverage consumption). Although high-sucrose ingestion increased power in the highest frequency band in the NTS in both lean and obese women, the groups differed in NTS functional connectivity patterns with obese subjects displaying significantly weaker NTS functional connectivity with a number of brain regions.

Some evidence for differential effects of sucrose ingestion on hypothalamus oscillatory dynamics were observed; in lean subjects, high-sucrose ingestion decreased power in the lowest frequency band for the hypothalamus, although it increased hypothalamus LF power in obese subjects. Although these changes in frequency power in the hypothalamus were

statistically a trend, group differences in right lateral hypothalamic functional connectivity were clearly significant with differential connectivity to reward and gustatory-related regions (discussed in the section on “Evidence for Altered Homeostatic and Reward Systems in Obese”). Finally, obese women, but not lean women, demonstrated a relationship between subjective taste ratings of the beverages and subsequent oscillatory dynamics of the hypothalamus and NTS, suggesting altered hedonic–homeostatic relationships in obese women. To our knowledge, this is the first study demonstrating the effect of sucrose ingestion on the intrinsic oscillation dynamics in several brain regions, playing a central role in the control of sucrose intake. In addition, the study suggests possible differences in brain mechanisms regulating subjective measures related to ingestive behavior.

Physiological Relevance of Changes in Frequency Spectrum of Intrinsic Brain Oscillations

In the current study, we examined region-selective changes in frequency power of intrinsic brain oscillations within multiple frequency bands as a way to monitor responses of the brain to nutrient ingestion. Intrinsic oscillations are known to arise at least in part from intrinsic neuronal activity.[25] Change in frequency power toward high frequencies is a finding that has been noted in several studies examining influences on the temporal dynamics of intrinsic BOLD activity using similar frequency bands to the current study, even though the neurophysiologic correlates of these frequency bands remain incompletely understood. For example, acute tryptophan depletion increases HF power in brainstem raphe nuclei and performance of a task has been shown to increase HF power in task-relevant regions, suggesting increased high frequency might represent increased neural activity.[26,27]

Greater Engagement of NTS After High-Sucrose Ingestion

The increase in NTS HF power after ingestion of sucrose could result from an increase in intrinsic neural activity due to vagal stimulation associated with activation of different glucose sensors at multiple levels of the gut–brain axis. Glucose-sensing mechanisms have been described in the tongue, intestine, pancreas, portal vein, hypothalamus, and NTS.[28] Glucose in the intestine results in the activation of enteroendocrine cells, release of incretin hormones and serotonin, and subsequent activation of vagal afferents.[29,30] Sugar and other sweeteners can activate lingual and intestinal sweet taste receptors without glucose absorption.[3,4,31] Activation of lingual G-protein–coupled receptors of the T1R family that mediate sweet taste in the mouth will increase neuronal activity in the rostral NTS via activation of sensory neurons in the chorda tympani. In contrast, evidence for the activation of the vagal afferent pathways by ligands of intestinal sweet taste receptors on enteroendocrine cells has not been reported, and such activation can only produce local changes in gastrointestinal function.[32] In contrast to lingual and intestinal sweet taste receptors, other glucose-sensing pathways, such as adenosine triphosphate–activated K channels in the endocrine pancreas and intestine, depend on absorption and metabolism of glucose, and will be differentially activated by nutritive and non-nutritive sweeteners.[5,6] As we compared brain responses between 2 drinks of similar sweetness but significantly different sucrose content, one might speculate that the greater HF power in the NTS after the high-sucrose drink was related primarily to differences in the activation of intestinal vagal afferents by mechanisms other than sweet taste receptor activation. However, alternative

explanations, including greater gastric vagal stimulation by a slower emptying of the high-sucrose drink cannot be ruled out.[33]

In addition to the hypothesized greater activation of intestinal vagal afferents by absorbed glucose, it is also possible that an increase in plasma glucose associated with the high-sucrose drink might stimulate glucose sensors in the portal vein,[34] or on central neurons, including those in the NTS. However, the fact that the increase in HF power during the high-sucrose condition was seen as early as 10–15 minutes after beverage consumption, when peripheral blood glucose and GLP-1 levels are most likely just beginning to rise,[35] suggests that this effect does not depend on near-maximal levels of peripheral blood glucose or GLP-1 and might reflect more direct enteric-central nervous system communication. Activation of vagal afferents after oral glucose has been shown in rats to occur relatively quickly, within 5–6 minutes.[36]

Evidence for Altered Homeostatic and Reward Systems in Obese

Although the low- and high-sucrose beverages did not differ in terms of subjective taste, obese women rated the taste of both beverages lower than lean women, consistent with a reduced hedonic response. For obese women, but not lean women, these taste ratings were negatively associated with subsequent intrinsic oscillatory dynamics of 2 homeostatic regions (NTS and hypothalamus), eg, more positive taste ratings correlated with less HF power in the NTS, and with less LF power in the right lateral hypothalamus, suggesting a connection between reduced hedonic experience and homeostatic activity not seen in lean women.

Although HF power in the NTS increased after high-sucrose ingestion for both lean and obese women, the functional connectivity of the NTS with other brain regions at the same oscillation frequency differed between the groups. Lean subjects demonstrated greater connectivity of the NTS and PCC, a major node of the default mode network involved in self-related processes.[37] Engagement of the PCC has been observed during the cognitive reappraisal of food.[38] Although the precise behavioral correlate of synchronized oscillation within different brain regions remains to be determined, one may speculate that the greater synchrony of HF oscillations in a homeostatic input region (NTS) and the PCC reflect greater incorporation of homeostatic input signals in the perception of food relevance to self, in lean women compared with obese women.

The functional connectivity of the right lateral hypothalamus within the LF domain differed between the groups, especially after high-sucrose ingestion. The lateral hypothalamus is involved in the coordination of homeostatic and reward systems.[1] Lean women showed greater positive functional connectivity between the lateral hypothalamus and the posterior insula, which is the primary viscerosensory cortex and part of the homeostatic afferent brain network.[39] In addition, greater lateral hypothalamus functional connectivity in lean women was observed with BA43, a region associated with processing oral somatosensory aspects of gustatory stimuli.[40] In contrast, obese women demonstrated greater functional connectivity of the right lateral hypothalamus with the putamen. The putamen is part of the dorsal striatum and is involved in reward-related functions, including those related to ingestive behavior, such as approach or avoidance of food.[41] These results suggest differential

coordination between lateral hypothalamus LF oscillations and oscillations within the homeostatic and reward systems, such that obese women have enhanced coordination with a reward region and weaker coordination with homeostatic and gustatory regions compared with lean women.

Limitations of the study

The separation of frequency bands used in this analysis was based on the observation that behaviorally relevant brain oscillations have linearly distributed center frequencies on the natural logarithmic scale.[8] However, little is known about the functional relevance of these bands. Given that activation of brain regions through a task has been shown to shift frequency distribution toward higher frequencies,[42] one possible interpretation of the increased HF power in the NTS is that neural activity in this brain region is increased; however, other possibilities exist. The BOLD signal within a brain voxel combines signals from multiple neuronal populations, and it is possible that the contribution of the signal from a particular population is enhanced during sucrose ingestion. Cardiovascular and other physiological artifacts could potentially be affected by carbohydrate ingestion and also affect spectral properties. Cardiac-related pulsatile brainstem movements and blood flow could affect BOLD signals at or in multiples of the heart rate. Even though autonomic parameters were not collected, the current study minimized the impact of physiological noise by regression of cerebral spinal fluid and white matter signals before filtering and calculation of frequency power.[43] Also, physiological noise is minimal in slow-4 and slow-5 frequency bands, and is strongest in slow-2, a band not used in this study.[7]

The current study was not designed to define the relative roles of various mechanisms underlying glucose-induced gut to brain signaling, including the role of blood glucose, incretins, and insulin. For example, as we only examined brain activity occurring 10–20 minutes after beverage consumption, future studies are needed to provide a greater temporal profile of brain activity during nutrient metabolism and correlations of these profiles with endocrine and neurocrine mechanisms. In addition, the sample size of the current study (and of the pilot test of beverage similarity) was small, and the results need to be replicated in a larger sample. Finally, the hypothalamus demonstrated changes in a very small spatial extent. As the hypothalamus is a small structure consisting of multiple nuclei, it is possible that the results reflect alterations in a particular nucleus. The robustness of the results will need to be demonstrated with future studies.

Previous investigations of the impact of food intake on brain processes have typically employed task fMRI, in which the brain's response to visual food cues in the context of different physiological states are compared. In contrast, examination of intrinsic fluctuations in the BOLD signal provides an opportunity to study neurobiologic features associated with the physiologic state itself without the use of external cues. Also, the most popular designs compare ingestion of a meal with fasting or compare ingestion of glucose with water. These conditions differ in terms of sweet taste receptor activation, as well as vagal stimulation, due to the absorption of nutrients within the gut. The current study employed a design that better isolates the effect of vagal stimulation due to nutrient absorption on intrinsic brain activity. This study demonstrates effects of a high-calorie sucrose drink on key regions involved in

food homeostasis (NTS and other brain stem regions). In addition, it revealed differences in functional connectivity between lean and obese subjects providing support for altered interaction between homeostatic and reward networks in obese individuals.

To our knowledge, this is the first use of this approach to identify brain-circuit abnormalities related to ingestive behavior in obese subjects. The findings demonstrate the feasibility of examining intrinsic BOLD activity to study central effects of food intake and to identify differences between normal-weight and obese individuals. Frequency-based analysis of intrinsic activity and connectivity appears to be a sensitive, simple measure of the brain's response to nutrient ingestion, and might be useful as a biomarker in the study of obesity.

Acknowledgments

Funding

Supported by National Institutes of Health grants P30 DK041301, T32 DK007180, R24 AT002681, R01 DK048351, and P50DK64531.

Abbreviations used in this paper

BOLD	blood-oxygen level–dependent
fMRI	functional magnetic resonance imaging
HF	high frequency
LF	low frequency
MF	medium frequency
NTS	nucleus tractus solitaries
PCC	posterior cingulate cortex
PLS	partial least squares
VAS	visual analog scale

References

1. Volkow ND, Wang GJ, Baler RD. Reward, dopamine and the control of food intake: implications for obesity. Trends Cogn Sci. 2011; 15:37–46. [PubMed: 21109477]
2. Carnell S, Gibson C, Benson L, et al. Neuroimaging and obesity: current knowledge and future directions. Obes Rev. 2011; 13:43–56. [PubMed: 21902800]
3. Sternini C, Anselmi L, Rozengurt E. Enteroendocrine cells: a site of 'taste' in gastrointestinal chemosensing. Curr Opin Endocrinol Diabetes Obes. 2008; 15:73–78. [PubMed: 18185066]
4. Pepino MY, Bourne C. Non-nutritive sweeteners, energy balance, and glucose homeostasis. Curr Opin Clin Nutr Metab Care. 2011; 14:391–395. [PubMed: 21505330]
5. Tolhurst G, Reimann F, Gribble FM. Nutritional regulation of glucagon-like peptide-1 secretion. J Physiol. 2009; 587:27–32. [PubMed: 19001044]
6. Reimann F. Molecular mechanisms underlying nutrient detection by incretin-secreting cells. Int Dairy J. 2010; 20:236–242. [PubMed: 20204054]
7. Zuo XN, Di Martino A, Kelly C, et al. The oscillating brain: complex and reliable. Neuroimage. 2010; 49:1432–1445. [PubMed: 19782143]

8. Buzsáki G, Draguhn A. Neuronal oscillations in cortical networks. Science. 2004; 304:1926–1929. [PubMed: 15218136]
9. Zhang J, Wei L, Hu X, et al. Specific frequency band of amplitude low-frequency fluctuation predicts Parkinson's disease. Behav Brain Res. 2013; 252:18–23. [PubMed: 23727173]
10. Yu R, Hsieh MH, Wang HL, et al. Frequency dependent alterations in regional homogeneity of baseline brain activity in schizophrenia. PLoS One. 2013; 8:e57516. [PubMed: 23483911]
11. Han Y, Wang J, Zhao Z, et al. Frequency-dependent changes in the amplitude of low-frequency fluctuations in amnestic mild cognitive impairment: a resting-state fMRI study. Neuroimage. 2011; 55:287–295. [PubMed: 21118724]
12. Di Martino A, Ghaffari M, Curchack J, et al. Decomposing intra-subject variability in children with attention-deficit/hyperactivity disorder. Biol Psychiatry. 2008; 64:607–614. [PubMed: 18423424]
13. Zigmond As SRP. The hospital anxiety and depression scale. Acta Psychiatr Scand. 1983; 67:361–370. [PubMed: 6880820]
14. Flint A, Raben A, Blundell JE, et al. Reproducibility, power and validity of visual analogue scales in assessment of appetite sensations in single test meal studies. Int J Obes Relat Metab Disord. 2000; 24:38–48. [PubMed: 10702749]
15. Connolly L, Coveleskie K, Kilpatrick LA, et al. Differences in brain responses between lean and obese women to a sweetened drink. Neurogastroenterol Motil. 2013; 25:579–e460. [PubMed: 23566308]
16. Chao-Gan Y, Yu-Feng Z. DPARSF: A MATLAB Toolbox for "Pipeline" Data Analysis of Resting-State fMRI. Front Syst Neurosci. 2010; 4:13. [PubMed: 20577591]
17. Song XW, Dong ZY, Long XY, et al. REST: a toolkit for resting-state functional magnetic resonance imaging data processing. PLoS One. 2011; 6:e25031. [PubMed: 21949842]
18. Zou QH, Zhu CZ, Yang Y, et al. An improved approach to detection of amplitude of low-frequency fluctuation (ALFF) for resting-state fMRI: fractional ALFF. J Neurosci Methods. 2008; 172:137–141. [PubMed: 18501969]
19. Maldjian JA, Laurienti PJ, Kraft RA, et al. An automated method for neuroanatomic and cytoarchitectonic atlas-based interrogation of fMRI data sets. Neuroimage. 2003; 19:1233–1239. [PubMed: 12880848]
20. Brett, M.; Anton, JL.; Valabregue, R., et al. Region of interest analysis using an SPM toolbox; Sendai, Japan. Eighth International Conference on Functional Mapping of the Human Brain; 2002;
21. Cole DM, Smith SM, Beckmann CF. Advances and pitfalls in the analysis and interpretation of resting-state FMRI data. Front Syst Neurosci. 2010; 4:8. [PubMed: 20407579]
22. McIntosh AR, Bookstein FL, Haxby JV, et al. Spatial pattern analysis of functional brain images using partial least squares. Neuroimage. 1996; 3:143–157. [PubMed: 9345485]
23. Hamlett, A.; Ryan, L.; Wolfinger, R. On the use of PROC MIXED to estimate correlation in the presence of repeated measures. SAS Users Group International, Proceedings of the Statistics and Data Analysis Section 2004, paper 198-29; p. 1-7.
24. Berthoud HR. Vagal and hormonal gut-brain communication: from satiation to satisfaction. Neurogastroenterol Motil. 2008; 20(suppl 1):64–72. [PubMed: 18402643]
25. Pan WJ, Thompson GJ, Magnuson ME, et al. Infraslow LFP correlates to resting-state fMRI BOLD signals. Neuroimage. 2013; 74:288–297. [PubMed: 23481462]
26. Salomon RM, Cowan RL, Rogers BP, et al. Time series fMRI measures detect changes in pontine raphe following acute tryptophan depletion. Psychiatry Res. 2011; 191:112–121. [PubMed: 21236648]
27. Baria AT, Baliki MN, Parrish T, et al. Anatomical and functional assemblies of brain BOLD oscillations. J Neurosci. 2011; 31:7910–7919. [PubMed: 21613505]
28. Heijboer AC, Pijl H, Van den Hoek AM, et al. Gut-brain axis: regulation of glucose metabolism. J Neuroendocrinol. 2006; 18:883–894. [PubMed: 17076764]
29. Raybould HE, Cooke HJ, Christofi FL. Sensory mechanisms: transmitters, modulators and reflexes. Neurogastroenterol Motil. 2004; 16(suppl 1):60–63. [PubMed: 15066007]

30. Racke K, Reimann A, Schworer H, et al. Regulation of 5-HT release from enterochromaffin cells. Behav Brain Res. 1996; 73:83–87. [PubMed: 8788482]

31. Young RL. Sensing via intestinal sweet taste pathways. Front Neurosci. 2011; 5:23. [PubMed: 21519398]

32. Jang HJ, Kokrashvili Z, Theodorakis MJ, et al. Gut-expressed gustducin and taste receptors regulate secretion of glucagon-like peptide-1. Proc Natl Acad Sci U S A. 2007; 104:15069–15074. [PubMed: 17724330]

33. Ma J, Bellon M, Wishart JM, et al. Effect of the artificial sweetener, sucralose, on gastric emptying and incretin hormone release in healthy subjects. Am J Physiol Gastrointest Liver Physiol. 2009; 296:G735–G739. [PubMed: 19221011]

34. Matveyenko AV, Donovan CM. Metabolic sensors mediate hypoglycemic detection at the portal vein. Diabetes. 2006; 55:1276–1282. [PubMed: 16644683]

35. Moller JB, Jusko WJ, Gao W, et al. Mechanism-based population modelling for assessment of L-cell function based on total GLP-1 response following an oral glucose tolerance test. J Pharmacokinet Pharmacodyn. 2011; 38:713–725. [PubMed: 21922329]

36. Vincent KM, Sharp JW, Raybould HE. Intestinal glucose-induced calcium-calmodulin kinase signaling in the gut-brain axis in awake rats. Neurogastroenterol Motil. 2011; 23:e282–e293. [PubMed: 21303432]

37. Fransson P, Marrelec G. The precuneus/posterior cingulate cortex plays a pivotal role in the default mode network: evidence from a partial correlation network analysis. Neuroimage. 2008; 42:1178–1184. [PubMed: 18598773]

38. Yokum S, Stice E. Cognitive regulation of food craving: effects of three cognitive reappraisal strategies on neural response to palatable foods. Int J Obes (Lond). 2013; 37:1565–1570. [PubMed: 23567923]

39. Craig AD. Interoception: the sense of the physiological condition of the body. Curr Opin Neurobiol. 2003; 13:500–505. [PubMed: 12965300]

40. Zald DH, Pardo JV. Cortical activation induced by intraoral stimulation with water in humans. Chem Senses. 2000; 25:267–275. [PubMed: 10866985]

41. Kaye WH, Fudge JL, Paulus M. New insights into symptoms and neurocircuit function of anorexia nervosa. Nat Rev Neurosci. 2009; 10:573–584. [PubMed: 19603056]

42. Baliki, M.; Baria, A.; Apkarain, AV. The cortical rhythms of chronic back pain. Quebec City, Canada: Organization for Human Brain Mapping; 2011.

43. Hallquist MN, Hwang K, Luna B. The nuisance of nuisance regression: spectral misspecification in a common approach to resting-state fMRI preprocessing reintroduces noise and obscures functional connectivity. Neuroimage. 2013; 82:208–225. [PubMed: 23747457]

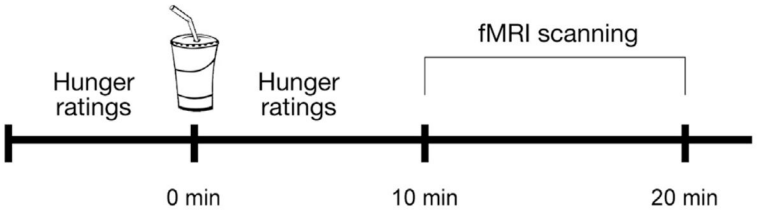

Figure 1.

Summary of procedures is given. On each scanning day, subjects completed hunger ratings immediately before and after beverage consumption (high or low sucrose). Commencing 10 minutes after consumption, subjects were scanned while resting for an additional 10 minutes.

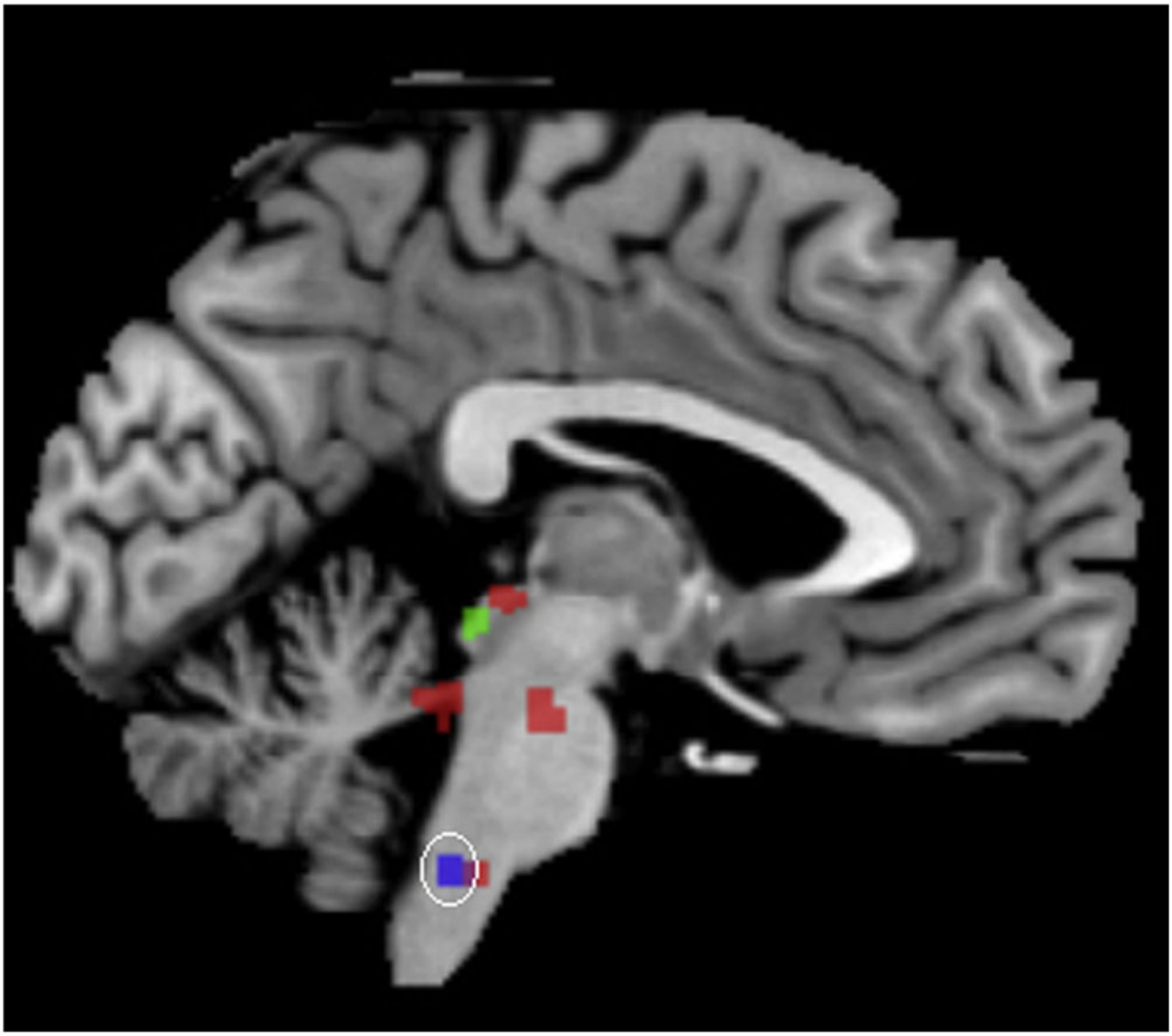

Figure 2.

Activity in brainstem regions demonstrated sucrose-related redistribution of frequency power. *Red*: reduced LF/MF frequency power after ingestion of a high-sucrose beverage compared with a low-sucrose beverage in both lean and obese women. *Blue*: greater HF frequency power after ingestion of a high-sucrose beverage compared with a low-sucrose beverage in both lean and obese women. *Green*: greater HF frequency power in lean compared with obese women regardless of drink condition. NTS is shown *circled*. frequency for lean women compared with obese women (Figure 2).

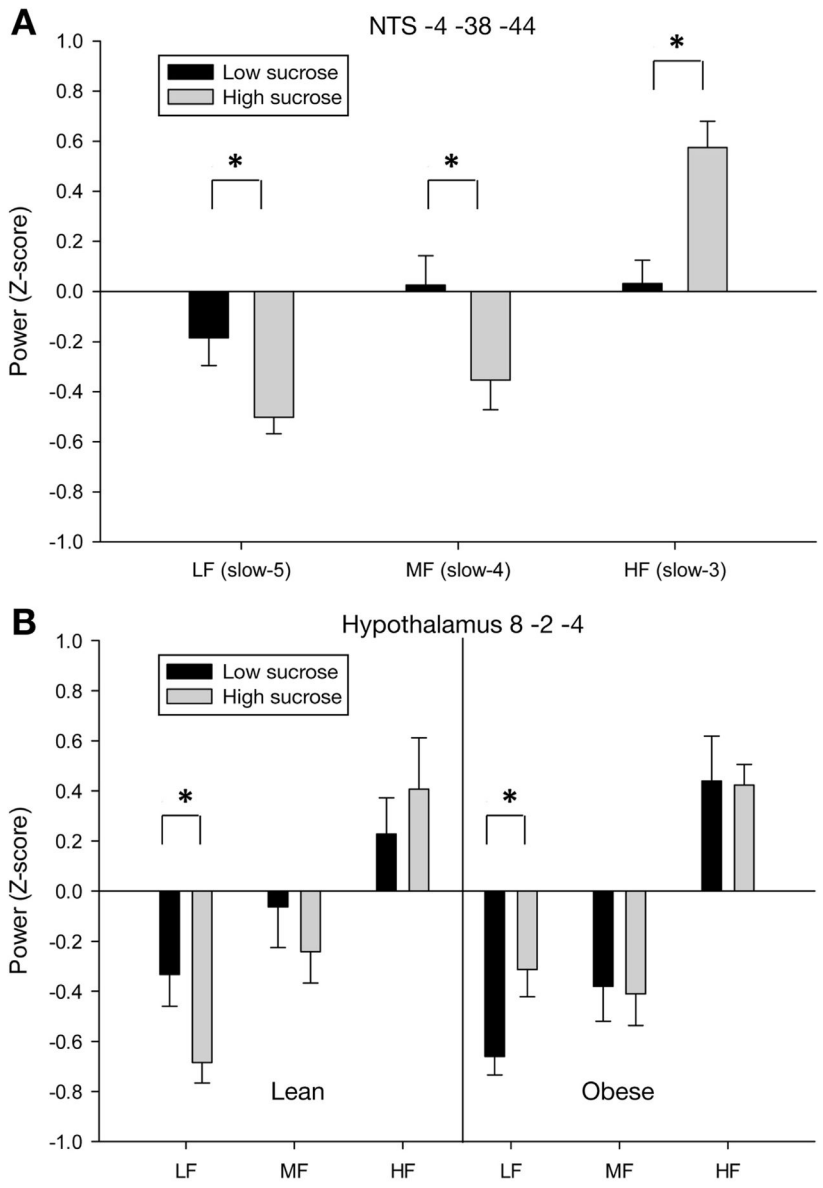

Figure 3.

Power within each frequency band is graphed for NTS (*A*) and hypothalamus (*B*).

Table 1

Means and Standard Errors for Age, Body Mass Index, Symptom, Taste, and Appetite Ratings (Before and After Beverage Consumption)

	Lean		Obese	
Age	24.91 (1.24)		27.64 (1.87)	
BMI	22.39 (0.45)		32.82 (0.68)[a]	
Depression	1.09 (.37)		2.36 (.43)[a]	
Anxiety	5.45 (1.16)		4.76 (.98)	
	Low sucrose	High sucrose	Low sucrose	High sucrose
Taste	8.18 (.39)	7.73 (.50)	6.86(.61)[a]	5.73 (.71)[a]
Hunger				
Before	5.46 (.55)	5.59 (.81)	5.50(.54)	5.68 (.44)
After	4.59 (.77)	4.09 (.72)[b]	4.91(.50)	5.27 (.49)
Satisfaction				
Before	1.46 (.37)	1.46 (.31)	2.46(.52)	2.09 (.49)
After	3.41 (.60)[b]	3.41(.58)[b]	3.50(.42)[b]	4.14 (.50)[b]
Desire for sweetness				
Before	5.32 (.66)	6.82 (.96)	4.59(.59)	5.41 (.69)
After	4.14 (.74)	3.14 (.65)[b]	2.64(.32)[b]	3.32 (.74)[b]

NOTE. Values are mean (SE).

BMI, body mass index (calculated as kg/m^2).

[a] $P < .05$ between group comparison.

[b] $P < .05$ within group comparison.

Table 2

Peak Coordinates (Montreal Neurological Institute) of Brain Regions Demonstrating Significant Sucrose or Group-Related Differences in Temporal Dynamics

Region	Contrast	Hemisphere	X	Y	Z	T	*P* value[a]	*P*(q)[b]	Cluster size
Slow-5									
Pontine/trigeminal nuclei	Low>high sucrose	Left	−10	−24	−28	4.58	<.001	.005	73
LCC	Low>high sucrose	Bilateral	−2	−38	−20	4.24	.019	.032	28
PAG	Low>high sucrose	Bilateral	0	−30	−2	4.53	.017	.032	29
Hypothalamus	Interaction	Right	8	−2	−4	3.99	.019	.057	2
Slow-4									
Reticular nuclei	Low>high sucrose	Bilateral	0	−32	−48	3.98	.041	.047	19
Slow-3									
NTS	High>low sucrose	Bilateral	−4	−38	−44	4.85	.047	.047	19

LCC, locus coeruleus; PAG, periaqueductal gray.

[a] *P* value is corrected for small volume and familywise error.

[b] *P*(q) is further corrected by false discovery rate (FDR) due to multiple comparisons for the 3 bands.

Table 3

Peak Coordinates (Montreal Neurological Institute) of Brain Regions Demonstrating Significant Group-Related Differences in Functional Connectivity of the Hypothalamus and NTS

Region	Contrast	Hemisphere	X	Y	Z	BSR	*P* value	Cluster size
Slow-5 HYP functional connectivity across drink conditions								
pINS	Lean > obese	Right	38	−20	10	4.89	<.001	39
BA43	Lean > obese	Right	58	−6	16	4.32	<.001	74
Putamen	Obese > lean	Right	16	4	−4	5.26	<.001	85
Cerebellum	Obese > lean	Right	20	−76	−38	4.74	<.001	33
Slow-5 HYP functional connectivity during high sucrose								
pINS	Lean > obese	Right	38	−18	10	5.99	<.001	31
BA7	Lean > obese	Right	24	−72	38	5.84	<.001	62
BA43	Lean > obese	Right	56	−8	12	5.23	<.001	48
BA31	Lean > obese	Left	−20	−72	20	4.81	<.001	38
aINS	Lean > obese	Right	34	−14	20	4.72	<.001	33
BA40	Lean > obese	Left	−60	−28	28	4.58	<.001	43
BA6	Lean > obese	Left	−16	−24	80	4.23	<.001	33
Putamen	Obese > lean	Right	16	4	−2	6.31	<.001	32
BA10	Obese > lean	Right	16	54	22	5.98	<.001	44
Cerebellum	Obese > lean	Right	36	−46	−36	5.39	<.001	48
Cerebellum	Obese > lean	Right	32	−90	−30	5.35	<.001	69
Slow-3 NTS functional connectivity across drink conditions								
PCC (BA23)	Lean > obese	Right	8	−46	24	5.64	<.001	78
Cerebellum	Lean > obese	Left	−28	−70	−28	5.31	<.001	65
Cerebellum	Lean > obese	Right	16	−74	−22	4.56	<.001	40
PCC (BA30)	Lean > obese	Left	−12	−66	8	4.45	<.001	58
BA22	Lean > obese	Right	58	−10	−6	4.31	<.001	42
BA31	Lean > obese	Right	26	−64	22	4.28	<.001	32
PCC (BA30)	Lean > obese	Right	12	−58	6	3.82	.001	38
Slow-3 NTS functional connectivity during high sucrose								
BA7	Lean > obese	Left	−28	−54	66	4.96	<.001	39

Region	Contrast	Hemisphere	X	Y	Z	BSR	*P* value	Cluster size
BA8	Lean > obese	Bilateral	0	26	50	4.91	<.001	32
ACC (BA32)	Lean > obese	Right	24	2	44	4.52	<.001	30

ACC, anterior cingulate cortex; aINS, anterior insula; BSR, bootstrap ratio; HYP, hypothalamus; pINS, posterior insula.