FUNCTIONAL MAGNETIC RESONANCE IMAGING OF REWARD SYSTEM FUNCTIONING IN OBESE WOMEN

by

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FUNCTIONAL MAGNETIC RESONANCE IMAGING OF REWARD SYSTEM FUNCTIONING IN OBESE WOMEN

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ABSTRACT

Investigators have found that exaggerated reactivity to food cues, especially those associated with high-calorie foods, leads to hyperphagia and may be a risk factor for obesity. Obese individuals also appear to be less sensitive to interoceptive cues such as hunger and may rely more on exteroceptive cues such as the sight of foods to determine ingestive behavior. The increased motivational potency of foods and food cues driving relatively greater food intake in obese individuals appears to be mediated in part by a hyperactive reward system including the nucleus accumbens/ventral striatum (NAc), amygdala (AMYG), and orbitofrontal cortex (OFC). Using functional magnetic resonance imaging (fMRI), we investigated reward-related activation in response to pictures of high-calorie and low-calorie foods in 12 obese compared to 12 normal-weight women. We tested whether there were group differences in modulation of brain activation in response to food cues by (1) hunger and (2) the hedonic value and incentive salience of the foods. We used connectivity analysis to test a simple reward circuit including NAc, AMYG, and OFC to determine whether there were group differences in the functional interactions of regions in this network in response to food stimuli. Compared to controls, obese women exhibited greater activation in response to high-calorie food images in 10 of 13 reward-related
regions of interest, whereas controls displayed relatively greater activation in only one region. We also found that the obese individuals had relatively greater reward system activation predicted by foods’ incentive salience more than their hedonic value, especially for high-calorie foods. In addition, the obese group had relatively less hunger-modulated reward system activation in response to food images, especially low-calorie food images. Finally, compared to controls, the obese group had a relative deficiency in AMYG projections modulating activation in both OFC and NAc, but excessive influence of the OFC projections modulating activation in NAc. In obese individuals, foods appear to have heightened motivational value likely mediated by a hyperactive reward system, coupled with deficient hunger and emotional signals, which fail to adequately modulate reward-related activation in response to food cues, possibly promoting hyperphagia and obesity.
DEDICATION

This dissertation is dedicated to my family and friends, who provided me with invaluable support and perspective throughout graduate school. In particular, this project is dedicated to my perpetual educators about life, my parents, Suzy and Jim Stoeckel.
ACKNOWLEDGMENTS

This project is the result of tireless work by the numerous exceptional people I have been fortunate to work with throughout graduate school, many of whom I will not name because there are so many. Most importantly, I would like to thank Dr. Rosalyn Weller, my primary advisor and dissertation chair, who has devoted countless hours and boundless energy to my graduate training. This project would not have been possible without her patience, encouragement, and rigorous approach to science. I would also like to thank Dr. James Cox, who has helped me understand the methods and tools of science and how to think and write as a scientist. I am especially appreciative of the support and perspective provided by my graduate committee chair, Dr. Edwin Cook III. I am also grateful to the other members of my dissertation committee, Drs. Donald Twieg and David Allison, for agreeing to oversee this very important component of my graduate training. I would also like to express appreciation to Dr. Adrienne Lahti, who has encouraged me to think as an independent scientist and who has been a wonderful sounding board for professional ideas and personal issues. Mark Bolding has provided priceless assistance and technical expertise with commendable patience as I have learned fMRI and how to use its auxiliary tools. I am appreciative of the technical training and creative insights provided by Dr. Barry Horwitz. I also appreciate the technical input from Dr. Jieun Kim and her patience in teaching me about connectivity analysis. I would like to thank Janice Lambert for answering all of my dissertation-related questions and ensuring that I had completed all the necessary paperwork on time. I am also grateful to David White and Shirley Townsend for their technical help and mental health check-ins during the preparation of the dissertation manuscript. I would like to thank Casie Tavares, who has provided me with unconditional emotional support and has shown me an often saintly amount of patience and understanding throughout this dissertation project. Finally, I would like to thank all of my family and friends, who have provided me with support and understanding throughout this often personally and professionally challenging process.

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<tr>
<td>3T</td>
<td>3 Tesla</td>
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<tr>
<td>ACC</td>
<td>anterior cingulate cortex</td>
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<td>AMYG</td>
<td>amygdala</td>
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<tr>
<td>ANOVA</td>
<td>analysis of variance</td>
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<td>BA</td>
<td>Brodmann area</td>
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<tr>
<td>BMI</td>
<td>Body Mass Index</td>
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<td>EPI</td>
<td>echo planar imaging</td>
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<tr>
<td>fMRI</td>
<td>functional magnetic resonance imaging</td>
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<td>FOV</td>
<td>field of view</td>
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<tr>
<td>GLM</td>
<td>General Linear Model</td>
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<tr>
<td>HC</td>
<td>high-calorie foods</td>
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<tr>
<td>HCSV</td>
<td>high-calorie (savory) foods</td>
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<tr>
<td>HCSW</td>
<td>high-calorie (sweet) foods</td>
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<tr>
<td>HIPPO</td>
<td>hippocampus</td>
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<td>HYPO</td>
<td>hypothalamus</td>
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<tr>
<td>Lat</td>
<td>lateral</td>
</tr>
<tr>
<td>LC</td>
<td>low-calorie foods</td>
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<tr>
<td>Med</td>
<td>medial</td>
</tr>
<tr>
<td>MNI</td>
<td>Montreal Neurological Institute</td>
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<td>NAc</td>
<td>nucleus accumbens</td>
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<td>Abbreviation</td>
<td>Description</td>
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<tr>
<td>OFC</td>
<td>orbitofrontal cortex</td>
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<td>PET</td>
<td>positron emission tomography</td>
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<td>PFC</td>
<td>prefrontal cortex</td>
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<td>PCA</td>
<td>principal components analysis</td>
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<td>ROI</td>
<td>region of interest</td>
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<tr>
<td>SE</td>
<td>standard error</td>
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<td>SPM</td>
<td>Statistical Parametric Mapping</td>
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<td>SVC</td>
<td>Small Volume Correction</td>
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<td>TE</td>
<td>echo time</td>
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<td>TR</td>
<td>repetition time</td>
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<tr>
<td>VAS</td>
<td>visual analog scale</td>
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<td>VTA</td>
<td>ventral tegmental area</td>
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INTRODUCTION

Obesity is a serious chronic health problem that has become an international epidemic, with an estimated 300 million clinically obese worldwide (WHO, 2003). The increase in the prevalence of obesity has most commonly been attributed to increased intake of calorically-dense foods high in sugar and saturated fats with low levels of nutrients, coupled with a decrease in physical activity, but as many as ten other factors have been implicated in this disease (WHO, 2003; Keith et al., 2006). Clearly, there is an urgent need to understand the underlying factors of obesity. Previous behavioral studies have suggested that exaggerated reactivity to food cues, especially those associated with high-reward foods (i.e., high-fat, energy dense foods), may be an etiologic factor in obesity (Mela, 2006). The heightened motivational potency of foods in obese individuals appears to be mediated in part by a hyperactive reward system (Del Parigi et al., 2003; James et al., 2004; Pelchat, 2002; Stoeckel et al., 2008; Volkow and Wise, 2005; Wang et al., 2004). In obese individuals, stimuli associated with highly rewarding foods may possess greater than normal potency for activating the reward system and, as a result, may trigger excessive motivation for non-homeostatic eating of such foods (Berthoud, 2004; Mela, 2006). Studies have also indicated that obese individuals are less sensitive to interoceptive cues such as hunger and rely more on exteroceptive cues such as the sight of foods to determine ingestive behavior (Doucet et al., 2003;
Hetherington et al., 1996; Schachter, 1968; Tuomisto et al., 1998; Vincent et al., 2008; Wansink et al., 2007). It is thought that foods’ incentive salience (i.e., motivational value), as opposed to their hedonic value (i.e., pleasantness), and the neural system mediating drive play a prominent role in contributing to increased food intake and subsequent weight gain in some individuals with obesity. Finally, it is possible that not only greater activation of the reward system, but also differences in the interaction of regions in this network may contribute to the relatively increased motivational value of foods in obese individuals.

Only a small number of functional imaging studies have found abnormal patterns of brain activation in obese individuals in response to food cues. There has been only one functional imaging study investigating whether patterns of brain activation in response to high- and low-reward foods differ between obese compared to normal-weight individuals within the same study. There have been no neuroimaging studies investigating 1) if self-report measures of hedonic value and incentive salience of foods in response to food cues predict different neural responses in obese and normal-weight participants; 2) whether hunger modulates response to food cues in obese individuals differently than in normal-weight individuals; and 3) how brain regions in the reward system interact in response to high- and low-calorie foods cues and whether these interactions are different for obese and normal-weight individuals. The primary objective of this project was to use functional magnetic resonance imaging (fMRI) in a novel research design investigating differences in regions of the brain involved
in reward processing in response to high- and low-calorie visual food cues between obese and normal-weight women. We also wanted to determine whether reward-related brain activation was modulated by interoceptive (hunger) and exteroceptive (hedonic value and incentive salience) factors differently in obese and normal-weight individuals. In addition to testing whether there were group differences in the magnitude and extent of reward-system activation in response to food cues, we also wanted to test whether there were group differences related to how brain regions in a simple reward network interacted in response to high- and low-calorie food cues.

**Manuscript 1** -- In the study described here, we used fMRI to compare reward-related brain responses in obese and normal-weight women to pictures of foods. Foods were grouped into high- and low-calorie categories so we could investigate whether differences in brain activation by pictures of food between obese and normal-weight individuals are especially marked for high-calorie foods.

**Manuscript 2** -- The purpose of this study was to test how subjective interoceptive (hunger) and exteroceptive (hedonic value and incentive salience) factors modulate brain activation in the reward system in response to high- and low-calorie food images in obese and normal-weight women. We hypothesized that subjective pleasantness ratings ("liking") and appetite ratings ("wanting") of food images would predict different patterns of brain activation coding hedonic
value ("liking") and incentive salience ("wanting") in response to food images in reward-related areas. Obese participants were also expected to show greater regional differences compared to normal-weight controls in "wanting" rather than "liking" areas. In addition, we hypothesized that subjective hunger would modulate reward-related brain activation in response to foods more in the normal-weight than the obese group.

**Manuscript 3** -- In this study, we used fMRI and a two-stage path analysis plus General Linear Model (GLM) approach to investigate the interactions of key reward structures (nucleus accumbens - NAc, amygdala - AMYG, and orbitofrontal cortex - OFC) in a simple network to determine whether this network functions differently in obese and normal-weight individuals in response to high- and low-calorie food cues. We expected to find a number of altered effective connections in our obese group that might help explain why foods, especially high-calorie foods, have increased motivational potency for these individuals.
WIDESPREAD REWARD-SYSTEM ACTIVATION IN OBESE WOMEN IN RESPONSE TO PICTURES OF HIGH-CALORIE FOODS

by

LUKE E. STOECKEL, ROSALYN E. WELLER, EDWIN W. COOK III, DONALD B. TWIEG, ROBERT C. KNOWLTON, AND JAMES E. COX

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Abstract

Behavioral studies have suggested that exaggerated reactivity to food cues, especially those associated with high-calorie foods, may be a factor underlying obesity. This increased motivational potency of foods in obese individuals appears to be mediated in part by a hyperactive reward system. We used a Philips 3T magnet and fMRI to investigate activation of reward-system and associated brain structures in response to pictures of high-calorie and low-calorie foods in 12 obese compared to 12 normal-weight women. A regions of interest (ROI) analysis revealed that pictures of high-calorie foods produced significantly greater activation in the obese group compared to controls in medial and lateral orbitofrontal cortex, amygdala, nucleus accumbens/ventral striatum, medial prefrontal cortex, insula, anterior cingulate cortex, ventral pallidum, caudate, putamen, and hippocampus. For the contrast of high-calorie vs. low-calorie foods, the obese group also exhibited a larger difference than the controls did in all of the same regions of interest except for the putamen. Within-group contrasts revealed that pictures of high-calorie foods uniformly stimulated more activation than low-calorie foods did in the obese group. By contrast, in the control group, greater activation by high-calorie foods was seen only in dorsal caudate, whereas low-calorie foods were more effective than high-calorie foods in the lateral orbitofrontal cortex, medial prefrontal cortex, and anterior cingulate cortex. In summary, compared to normal-weight controls, obese women exhibited greater activation in response to pictures of high-calorie foods in a large number of regions hypothesized to mediate motivational effects of food cues.
Keywords: food cues, high-calorie food, obesity, reward system
Recently, there has been increased interest in potential parallels between drug addiction and obesity (Del Parigi et al., 2003; Pelchat, 2002; Volkow & Wise, 2005). It is well established that rewarding effects of addictive drugs and natural reinforcers such as foods are mediated by a common neural substrate (Kelley and Berridge, 2002; Volkow and Wise, 2005; Wang et al., 2001; Wang et al., 2004b). Stimuli associated with primary rewards can acquire motivational potency such that they trigger “wanting” of the reward by virtue of their acquired ability to activate the reward system (Berridge, 2004). Current neural models of addiction have proposed that drug-related cues may trigger drug-seeking behavior by eliciting hyperactivity in a brain network of reward areas, such as the ventral tegmental area (VTA), nucleus accumbens/ventral striatum (NAc), amygdala (AMYG), orbitofrontal cortex (OFC), and ventral pallidum (Jentsch and Taylor, 1999; Kalivas and Volkow, 2005; Kolb, 1999; Robinson and Berridge, 2003; Simansky, 2005; Volkow et al., 2003, 2004). Food cues may play a similar role in the development and maintenance of obesity (Del Parigi et al., 2003; James et al., 2004; Pelchat, 2002; Volkow and Wise, 2005; Wang et al., 2004b). In obese individuals, stimuli associated with high-incentive foods, such as those high in fat and energy density, may possess greater than normal potency for activating the reward system and, as a result, triggering excessive motivation for non-homeostatic eating of such foods (Berthoud, 2004; Mela, 2006).

Preliminary neuroimaging studies have found differences in responses to food or food cues in obese subjects compared to controls (e.g., Del Parigi et al., 2004; Gautier et al., 2000; Matsuda et al., 1999). For example, PET studies have
found that consumption of a small (2 ml) quantity of liquid food produced greater increases in blood flow in several areas, such as insula and ventral midbrain, and greater decreases in medial orbitofrontal cortex (OFC) in obese subjects (Del Parigi et al., 2005; Karhunen et al., 1997). Most relevant to the present research, a recent functional magnetic resonance imaging (fMRI) study by Rothemund et al. (2007) compared activation in obese and normal-weight individuals in response to pictures of high- and low-calorie foods, eating-related utensils, and neutral control stimuli. These authors found that activation in the caudate, putamen, anterior insula, hippocampus, and parietal lobule was greater in the obese group than in controls for the contrast of high-calorie foods vs. neutral control stimuli. However, for several reasons this study may have missed additional brain regions differentially activated in their obese and normal-weight comparison groups. First, both groups rated the low-calorie food images higher in appeal than the high-calorie food images. Second, there may have been habituation effects, as have been observed in the amygdala (e.g., Holsen et al., 2005), because the same images were apparently shown more than once. Third, statistical power may have been low because a small number of brain volumes were collected per visual image condition. In addition, female subjects were not matched on stage of the menstrual cycle. This may have added variability and, therefore, reduced statistical power because reward- and emotion-related brain activation varies across the menstrual cycle (Amin et al., 2006; Dreher et al., 2007; Goldstein et al., 2005; Protopopescu et al., 2005).
In the study described here, we used fMRI to compare brain responses in obese and normal-weight women to pictures of foods. Foods were blocked into high- and low-calorie categories to allow us to investigate whether differences in brain activation by pictures of food between obese and normal-weight individuals are especially marked for high-calorie foods. Participants were matched on phase of the menstrual cycle as well as education, which might influence food preferences or familiarity. To minimize habituation, no image was used more than once. Our regions of interest (ROI’s) included sites within the reward system proper (AMYG, NAc, OFC, ventral pallidum, and VTA) as well as associated areas thought to be involved in motivational processes: anterior cingulate cortex (ACC), caudate nucleus, putamen, hippocampus (HIPPO), hypothalamus (HYPO), insula, and medial prefrontal cortex (Med PFC).

Materials and methods

Participants

Participants were 24 obese and normal-weight women recruited from the University of Alabama at Birmingham (UAB) area. Using the Body Mass Index (BMI), a proxy measure of obesity based on height and weight, 12 participants were classified as obese (BMI = 30.8 – 41.2) and 12 were normal-weight (BMI = 19.7 – 24.5). There were no group differences on mean age (obese: 27.8, SD = 6.2; control: 28.0, SD = 4.4), ethnicity (obese: 7 African-American, 5 Caucasian; control: 6 African-American, 6 Caucasian), education (obese: 16.7 years, SD = 2.2; control: 17.2, SD = 2.8), or mean day of the follicular phase of the menstrual
cycle (obese: day 6.8, SD = 3.1, control: day 5.7, SD = 3.3). Participants were recruited from the university community with newspaper advertisements and fliers. They were informed that the purpose of the study was to look at patterns of brain activity in hungry participants of different BMI’s in response to visual images of various objects such as foods and control images. To reduce variability among participants in their responses to the experimental stimuli, individuals were excluded based on the following criteria: a) a positive eating disorder history (Eating Disorder Diagnostic Scale, EDDS; Stice et al., 2000, 2004) because such individuals may feel disgust when viewing food images (Uher et al., 2004), b) food preferences that were inconsistent with our food images (e.g., vegetarian), c) current efforts to lose weight (e.g., dieting or participating in a weight-loss program), and d) irregular eating habits (not eating meals at typical times or not eating breakfast). Participants were also excluded based on vision poorer than 20/40 without glasses (Snellen eye chart), left-handedness (Edinburgh Handedness Inventory; Oldfield, 1971), cigarette smoking, claustrophobia, pregnancy, lactation, irregular menstrual cycles, a chronic health condition such as diabetes or hypertension, current or past drug or alcohol abuse problem, current psychoactive medication use, positive history of psychosis, history of head injury or other event resulting in loss of consciousness exceeding two minutes, other neurological impairment, hearing impairment requiring a hearing-aid, or ferromagnetic material in the body. In addition, our scanner required that we limit participation to those with weights < 305 pounds (138 kg) with girth < 64 inches (163 cm).
Stimuli

The stimuli consisted of 252 color pictures either selected from a previous study (Stoeckel et al., 2007) or taken with a digital camera, scanned, and modified for consistent size, resolution, and luminance (see Stoeckel et al., 2007, for further details). The 168 food images were subdivided into low-calorie and high-calorie categories, each consisting of 84 unique images. Low-calorie food images consisted of such low-fat items as steamed vegetables and broiled fish. High-calorie foods were primarily items high in fat. Women have been reported to prefer sweet foods with a high fat content, as opposed to savory foods with a high fat content (Drewnowski et al., 1992; Wansink et al., 2003), so we further subdivided our 84 high-calorie food images into sweet and savory foods. The 42 images of high-calorie sweet foods were desserts such as cheesecake or breakfasts such as pancakes with syrup. The 42 high-calorie savory foods included such items as fatty meats (e.g., ribs) and snacks (e.g., cheese nachos). Control stimuli consisted of car images because we thought it important that the control images be moderately emotionally engaging to help maintain participants’ attention while in the magnet. Although fMRI studies have reported that images of desirable sports cars or desirable car logos activate reward system structures (Erk et al., 2003; Schafer et al., 2006), we used a wide range of car models and ages and found that food images were generally more effective in activating our ROI’s than the car images were.
Procedure

After phone screening, informed consent, and in-person screening to validate BMI, eating disorder diagnosis, and other study criteria, participants were scheduled for the fMRI session. They were instructed to eat a normal breakfast between 7-8 A.M. but to skip lunch and consume only water so that they had fasted for approximately 8-9 h before being imaged between 3-5 P.M. Before being imaged, each participant took a urine-based pregnancy test at the UAB Pittman General Clinical Research Center (GCRC) to confirm that she was not pregnant, as required for fMRI research by our Institutional Review Board. At that time, the participant’s height and weight were recorded. Participants were asked to provide subjective hunger ratings using three visual analog scales immediately prior to the imaging session to validate the fasting manipulation. The scales consisted of 150 mm lines with the left ends anchored at “none at all” and the right at “extremely” or “most possible” (Nolan et al., 2003; Stoeckel et al., 2007). Hunger was assessed using the following three questions: 1) How hungry are you? 2) How strong is your desire to eat your favorite food now? 3) How much food would you like to eat right now?

While participants were in the magnet, visual stimuli were presented in a block design format, with a total of six 3:09 min runs per imaging session. Each run consisted of two epochs each of cars (C) and low-calorie foods (LC), and one epoch each of high-calorie sweet (HCSW) and high-calorie savory (HCSV) foods pseudorandomly presented to the participants (Fig. 1). Within each 21 s epoch of food or car images, seven individual images were presented for 2.5 s each
followed by a 0.5 s gap each, separated by 9 s of a gray blank screen with a fixation cross. Each run began and ended with 9 s of blank screen with the central fixation cross. Each run consisted of 63 volumes for a total of 378 volumes across six runs, of which 84 volumes were acquired during each of the car and low-calorie food exposures, and 42 volumes were acquired during each of the high-calorie sweet and savory food exposures. The visual images were presented by a laptop using VPM software (Cook et al., 1987), which was connected to a Sony LCD projector (VPLPX35) with a Navitar long-throw lens. Images were projected onto a screen beyond the participant’s head and viewed via a 45° single-surface rear-projecting mirror attached to the head coil. The LCD projector outside of the magnet room projected the image though a 8" diameter waveguide in the rear wall. The visual display was 10° x 13°.

![Fig. 1. One of the six runs used in the fMRI study. Within each run, six blocks of visual images, two each of cars, low-calorie foods (LC), and high-calorie foods (HC; one each of sweet, HCSV, and savory, HCSV), alternated with a gray blank screen with a fixation cross. Order of blocks was randomized from run to run with the constraint that a given stimulus category was not followed by the same category. Sec, seconds.](image-url)
Following the scans, subjects were given a recognition test consisting of 100 laminated color food and car images to sort into two piles, those seen in the magnet and those not seen. The recognition test was used to ensure that the obese and control groups did not differ systematically in their attention to and encoding of images while in the magnet. Participants also rated 50 food and car stimuli randomly selected from the imaging session on emotional valence (i.e., pleasantness; Osgood et al., 1957; Russell, 1979). Ratings for each image were manually recorded on 9-point Likert scales. A rating of 1 on the valence scale indicated an extremely unpleasant response to the visual image, a rating of 5 indicated a neutral response, and a rating of 9 indicated an extremely pleasant response to the visual image. The last six participants in the control group and last seven in the obese group completed the Positive and Negative Affect Scale (PANAS; Watson et al., 1988) in order to assess whether there were group differences on negative emotion, which has been shown to produce different brain activation patterns compared to positive emotion when individuals were shown high- and low-calorie visual food cues (Killgore and Yurgelun-Todd, 2006; 2007). Participants were financially compensated for their participation. All procedures were reviewed and approved by UAB’s Institutional Review Board for Human Use.

MRI acquisition and processing

Structural and functional MRI data were acquired using a Philips Intera 3T ultra-short bore magnet located within UAB’s Cardiovascular MRI facility. Each
session consisted of the following scans: a) scout (3-plane localizer scan); b) SENSE calibration scan; c) fMRI scans of activation to food and non-food images, and d) 3-D high-resolution anatomical scan. Head motion was restricted using memory foam inserts in the SENSE head coil. Functional MR images were acquired using a single-shot T2*-weighted gradient-echo EPI pulse sequence. We used TE = 30 msec, TR = 3 sec, and an 85º flip angle for 30 axial slices 4 mm thick with a 1 mm interslice gap, a scan resolution of 80 x 79, reconstructed to 128 x 128, and with a 230 x 149 x 230 mm FOV. The first four scans were discarded to allow the magnet to achieve steady-state magnetization. The start of each run of stimuli presentation was manually synchronized with MRI data acquisition. Although we did not monitor eye movements during imaging, emotional and neutral images have not been found to result in differential eye movements (Lane et al., 1999; Lang et al., 1998). For the high resolution anatomical scan, 160, 1 mm thick parasagittal 3-D slices were acquired using a T1 Turbo-field Echo (TFE) sequence with a SENSE factor of 2, T1 = 400 ms, TR = 9.9 ms, TE = 4.6 ms, 8º flip angle, and a scan resolution of 240 x 200, reconstructed to 256 x 256 for a FOV of 240 x 240 x 160.

Data were preprocessed using the SPM2 software package (Wellcome Dept. Imaging Neuroscience, London, UK). The fMRI data were realigned using INRIAlign, a motion correction algorithm unbiased by local signal changes that realigns to the first image volume collected (Freire et al., 2002). A mean functional image was computed. The mean EPI image was matched to the EPI template provided within SPM2. Data were then spatially transformed
(normalized) into standard MNI space using a customized algorithm with both linear and nonlinear components (Friston et al., 1995). This transformation was then applied to the corresponding functional images which were resliced into 2 x 2 x 2 mm resolution in MNI space. Normalized data were smoothed using a 6 mm FWHM Gaussian filter. No data sets failed to meet the movement inclusionary criteria, which were that movement before correction was < 2 mm in translational movement and < 2° in rotational movement.

Data analysis

Behavioral measures. Valence ratings of high- and low-calorie food images were analyzed with a mixed between-group (BMI group), within-group (Food Category) analysis of variance (ANOVA). Food categories were compared within each group using simple-effects analyses (Howell, 2007). A similar analysis was used to compare ratings of high-calorie sweet and high-calorie savory foods. Ratings on the three hunger scales were analyzed with a mixed between- (BMI group), within-group (scale) ANOVA. Comparisons of the groups on individual scales were performed with simple-effects analyses. Group means for numbers of correctly identified stimuli and false positives on the recognition test and negative affect scores from the PANAS were compared using independent-samples t tests.

fMRI data. Block-design blood oxygen level dependent (BOLD) responses were analyzed within the context of the General Linear Model on a voxel by voxel basis as implemented in SPM2 (Friston et al., 1995). The time course of brain activation was modeled with a boxcar function convolved with the canonical
hemodynamic response function (HRF) and a temporal derivative function. A first order autoregressive model was also implemented to correct for autocorrelations in the error term of the fMRI model.

A two-stage procedure was used for the statistical analysis of a mixed-effects design. At the first level (fixed effects), the fMRI data from each individual participant were used to generate statistical contrasts for comparing brain activation to 1) high-calorie foods vs. cars, 2) low-calorie foods vs. cars, 3) high-calorie foods vs. low-calorie foods, and 4) high-calorie sweet foods vs. high-calorie savory foods. These contrasts were then entered into a second level (random effects) analysis to compare the obese and normal-weight groups. For these statistical analyses, we used a region of interest (ROI) approach to address our a priori hypotheses and to improve statistical power. Our areas of interest were reward areas: VTA, NAc, AMYG, ventral pallidum, and OFC. We included both lateral (Lat) and medial (Med) OFC as ROI’s despite some signal drop-out that occurred in the latter because of strong magnetic susceptibility gradients between that portion of the brain and adjacent air spaces in the sinuses and nasal cavities (Ojemann et al., 1997). We also analyzed other, functionally associated regions that have been found to be activated in comparable fMRI and PET studies: ACC, Med PFC, insula, hippocampus (HIPPO), hypothalamus (HYPO), caudate and putamen. We then ran the analysis using small volume correction (SVC) within SPM2 to adjust for the number of voxels within our ROI’s. Significance for these a priori ROI’s was assessed at p < .01 with a spatial extent of seven voxels. Finally, to explore the possible significance of variability of BMI
within the obese group, that is, that higher BMI within this group might predict greater activation, we also performed a within-group (obese only) analysis using BMI as a regressor for the high-calorie vs. low-calorie contrast.

ROI's were defined using the WFU Pickatlas and the AAL and Talairach Daemon atlases (Lancaster et al., 2000; Maldjian et al., 2004; Tzourio-Mazoyer et al., 2002). Regions that were unavailable in these libraries (VTA and NAc) were drawn within the WFU Pickatlas using three-dimensional spheres centered at a voxel location that was determined by averaging voxel location dimensions from relevant fMRI studies (Aron et al., 2005; Menon and Levitin, 2005; O'Doherty et al., 2002). MNI coordinates for the voxels of maximum activation within an ROI obtained from SPM2 were entered into the WFU Pickatlas using the AAL and Talairach Daemon atlases to determine the anatomical location of each locus. In some instances, the AAL and Talairach Daemon atlases did not identify or were inconsistent in classifying a particular locus. Therefore, the classification of activated voxels was verified by visual inspection of the data, and unidentified or misclassified loci of activation were identified using a human brain atlas (Mai et al., 2004).

Results

Behavioral measures

There was no BMI-Group effect in an ANOVA comparing the groups on the aggregated hunger ratings ($F[1,22] = 0.84, p = .37$; Fig. 2), and there were no
group differences on the individual scales (all p values > .30). These outcomes suggest that our fMRI results did not result from group differences in hunger. Fig. 3 shows mean valence ratings of low calorie and high calorie food images by control and obese participants. ANOVA revealed a significant group by food category interaction (F[1,22] = 4.39, p = .048). A simple effects analysis showed that obese individuals’ ratings of high calorie foods were higher than their ratings of low calorie foods (F[1,22]=4.42, p = .047). Within the control group, ratings of high- and low-calorie foods were not significantly different (F[1,22] = 0.74, p = .40). ANOVA revealed no between- or within-group differences on ratings of the high-calorie sweet and high-calorie savory images (all p values > .40). The obese and control groups performed very similarly on the image recognition test performed after the scan, with mean percentages (±SEM) of images correctly identified of 65.7 ± 6.4% and 68.2 ± 4.1% for the control and obese participants, respectively. Both groups performed significantly above chance (p values < .05) and did not differ significantly (t[22] = 0.33, p = .74). Number of false positives also did not differ significantly (t[22] = 0.50, p = .62). Finally, based upon the partial samples who filled out the PANAS, there were no group differences in negative affect (t[11] = 0.39, p = .70).
Fig. 2. Mean hunger ratings (with SE) on the three VAS scales for the questions (1) How hungry are you?, (2) How strong is your desire to eat your favorite food now?, and (3) How much food would you like to eat right now?
Fig. 3. Mean valence ratings (with SE) of food images by obese (N=12) and control (N=12) participants. *P < 0.05 for obese group’s ratings of high-calorie vs. low-calorie foods, as well as the Category X Group interaction.

**fMRI results**

*Within-groups analyses: control group.* Within the control group, both high- and low-calorie food images were generally effective in activating cortical ROI’s relative to control images (e.g., Fig. 4A, dorsal Med PFC), with subcortical activation only in HIPPO (Table 1). Low-calorie foods produced more activation than high-calorie foods did in three ROI’s in frontal cortex (Lat OFC, Med PFC, and ACC). Only in dorsal caudate did high-calorie foods produce more activation than low-calorie foods.

*Within-groups analyses: obese group.* A within-group analysis of the obese subjects for the contrast of high-calorie foods vs. control images revealed significant activation in all of our ROI’s except for HYPO (Table 2; e.g., Fig. 4B,
VTA). Low-calorie food images activated the same ROI’s with the exception of Med OFC, VTA, and ventral pallidum. However, in six ROI’s, as listed in Table 2, low-calorie foods produced less activation than high-calorie foods. Low-calorie foods were not more effective than high-calorie foods in any ROI in the obese group. When BMI was used as a regressor for the high-calorie vs. low-calorie contrast, significant positive correlations were found for right Med OFC, right Med PFC, and right putamen (Supplementary Table 1).

It is noteworthy that, within the obese group, activation was stronger in response to sweet high-calorie foods than to savory high-calorie foods in ACC, insula, HIPPO, caudate, and putamen (Table 2; e.g., Fig. 4C, HIPPO; Fig 4D, Insula). A difference in the opposite direction was seen only in a cluster within the right insula.
Table 1.
Within-group comparisons for the control group contrasting differences in the high- and low-calorie food and car conditions.

<table>
<thead>
<tr>
<th>Contrast</th>
<th>ROI</th>
<th>Hem</th>
<th>Cluster</th>
<th>BA</th>
<th>t</th>
<th>MNI coordinates</th>
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For all contrasts (HiCal, high-calorie foods; LoCal, low-calorie foods), activated regions of interest (ROI) as identified by the WFU PickAtlas, right or left (R, L) hemisphere (Hem), number of contiguous voxels (Cluster), approximate Brodmann areas (BA) as identified in the WFU PickAtlas*, t-values, and coordinates of the voxel of greatest activation within the Montreal Neurological Institute (MNI) coordinate system are displayed. There were no significant activations for Sweet > Savory, Savory > Sweet. Other abbreviations as in the text.

* Brodmann's areas (BA) were identified in the WFU Pickatlas based on their anatomical location in Talairach space after converting the MNI coordinates to Talairach coordinates. As the locations of BA as defined in the Talairach Daemon are estimates based on an approximate conversion, the BA of some of the cortical foci in our study remained unidentified by the WFU Pickatlas.
Table 2.

Within-group comparisons for the obese group contrasting differences in the high- and low-calorie food and car conditions.

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<td>16, 28, 28</td>
<td></td>
</tr>
<tr>
<td>Insula</td>
<td>L</td>
<td>24</td>
<td>4.24</td>
<td>-30, -22, 22</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>R</td>
<td>11</td>
<td>3.90</td>
<td>46, 4, 2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HIPPO</td>
<td>L</td>
<td>7</td>
<td>3.05</td>
<td>-16, -8, -14</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>R</td>
<td>74</td>
<td>4.23</td>
<td>20, -10, -18</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caudate</td>
<td>L</td>
<td>8</td>
<td>3.69</td>
<td>-20, 16, 14</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>R</td>
<td>72</td>
<td>4.95</td>
<td>16, 16, 14</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Putamen</td>
<td>L</td>
<td>47</td>
<td>3.50</td>
<td>-22, 18, 0</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>R</td>
<td>61</td>
<td>4.01</td>
<td>32, 2, -8</td>
<td></td>
<td></td>
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<tr>
<td>Savory &gt; Sweet</td>
<td>Insula</td>
<td>R</td>
<td>35</td>
<td>3.41</td>
<td>40, -24, 4</td>
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</tr>
</tbody>
</table>

There were no significant activations for LoCal > HiCal. Conventions as in Table 1.
Fig. 4. A. Greater activation in controls to low-calorie foods > cars contrast in left dorsal Med PFC (sagittal view, cluster size = 22, Brodmann area, BA, 9, MNI coordinates: -8, 36, 30); B. Greater activation in obese participants to high-calorie foods > cars contrast in VTA (sagittal view, cluster size = 8, MNI coordinates: 4, -16, -10); C. Greater activation found in obese participants to sweet > savory high-calorie foods in right HIPPO (coronal view, cluster size = 74, MNI coordinates: 20, -10, -18); D. Greater activation found in obese participants to sweet > savory high-calorie foods in right insula (sagittal view, cluster size = 11, MNI coordinates: 46, 4, 2). Color bars indicate t-values. Activation is overlaid on the SPM2 single-subject T1 template.
Group comparisons: High-calorie food condition. As shown in Table 3, the contrast of high-calorie foods vs. cars was significantly greater in the obese group than in controls in all of the ROI’s but HYPO and VTA (e.g., Fig. 5A, ACC; Fig. 5B, insula; Fig. 5C, Lat OFC). Controls showed greater activation than the obese only in right Med PFC.

Table 3.

Between-group (obese vs. control group) comparisons contrasting differences in the high-calorie food > car and low-calorie food > car conditions.

<table>
<thead>
<tr>
<th>Contrast</th>
<th>ROI</th>
<th>Hem</th>
<th>Cluster</th>
<th>BA</th>
<th>t</th>
<th>MNI coordinates</th>
</tr>
</thead>
<tbody>
<tr>
<td>OB &gt; CTRL:</td>
<td>Med OFC</td>
<td>L</td>
<td>11</td>
<td></td>
<td>3.48</td>
<td>-20, 16, -14</td>
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<tr>
<td>HiCal &gt; Cars</td>
<td>Lat OFC</td>
<td>L</td>
<td>191</td>
<td></td>
<td>4.52</td>
<td>-32, 22, -18</td>
</tr>
<tr>
<td></td>
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<td>R</td>
<td>90</td>
<td>47</td>
<td>4.35</td>
<td>38, 20, -20</td>
</tr>
<tr>
<td></td>
<td>Med PFC</td>
<td>L</td>
<td>73</td>
<td>8</td>
<td>4.29</td>
<td>-2, 26, 44</td>
</tr>
<tr>
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<td>6</td>
<td>3.59</td>
<td>8, 28, 40</td>
</tr>
<tr>
<td></td>
<td>ACC</td>
<td>L</td>
<td>54</td>
<td></td>
<td>3.29</td>
<td>-12, 32, 20</td>
</tr>
<tr>
<td></td>
<td></td>
<td>R</td>
<td>30</td>
<td>24</td>
<td>3.34</td>
<td>8, 34, 16</td>
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<tr>
<td></td>
<td>Insula</td>
<td>L</td>
<td>26</td>
<td></td>
<td>3.62</td>
<td>-38, -14, -8</td>
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<tr>
<td></td>
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<td>R</td>
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<td></td>
<td>3.37</td>
<td>44, -2, 6</td>
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<td></td>
<td>NAc</td>
<td>R</td>
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<td>3.01</td>
<td>6, 14, -8</td>
</tr>
<tr>
<td></td>
<td>AMYG</td>
<td>L</td>
<td>12</td>
<td></td>
<td>3.27</td>
<td>-20, 0, -24</td>
</tr>
<tr>
<td></td>
<td>Vent Pall</td>
<td>L</td>
<td>10</td>
<td></td>
<td>3.53</td>
<td>-16, -2, 4</td>
</tr>
<tr>
<td></td>
<td>HIPPO</td>
<td>L</td>
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<td></td>
<td>3.70</td>
<td>32, -8, -26</td>
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<td></td>
<td>Caudate</td>
<td>R</td>
<td>11</td>
<td></td>
<td>3.12</td>
<td>16, 0, 18</td>
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<td></td>
<td>Putamen</td>
<td>L</td>
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<td></td>
<td>4.88</td>
<td>-22, -2, 12</td>
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<td>Contrast</td>
<td>ROI</td>
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<td>Cluster</td>
<td>BA</td>
<td>t</td>
<td>MNI coordinates</td>
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<tr>
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<td>---------</td>
<td>----</td>
<td>----</td>
<td>-----------------</td>
</tr>
<tr>
<td>CTRL &gt; OB: HiCal &gt; Cars</td>
<td>Med PFC</td>
<td>R</td>
<td>41</td>
<td>38</td>
<td>3.58</td>
<td>32, 2, 0</td>
</tr>
<tr>
<td>OB &gt; CTRL: LoCal &gt; Cars</td>
<td>Caudate</td>
<td>R</td>
<td>25</td>
<td>8</td>
<td>4.60</td>
<td>18, 60, 10</td>
</tr>
<tr>
<td></td>
<td>Putamen</td>
<td>L</td>
<td>27</td>
<td>18</td>
<td>2.85</td>
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<tr>
<td></td>
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<td>R</td>
<td>9</td>
<td>9</td>
<td>2.95</td>
<td>20, 16, -4</td>
</tr>
<tr>
<td>CTRL &gt; OB: LoCal &gt; Cars</td>
<td>Lat OFC</td>
<td>L</td>
<td>12</td>
<td>11</td>
<td>3.04</td>
<td>-26, 44, -12</td>
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<td>11</td>
<td>3.26</td>
<td>32, 42, -14</td>
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<tr>
<td></td>
<td>Med PFC</td>
<td>R</td>
<td>8</td>
<td>8</td>
<td>2.94</td>
<td>16, 60, 14</td>
</tr>
<tr>
<td></td>
<td>ACC</td>
<td>R</td>
<td>7</td>
<td>7</td>
<td>2.91</td>
<td>10, 44, 0</td>
</tr>
</tbody>
</table>

OB = obese group, CTRL = control group. Remaining conventions as in Table 1.
Fig. 5. Greater activation found in obese than in control participants to high-calorie foods > cars in (A) right anterior cingulate cortex (sagittal view, cluster size = 30, BA 24, MNI coordinates: -12, 32, 20), (B) left insula (sagittal view, cluster size = 26, MNI coordinates: -38, -14, -8), and (C) left lateral orbitofrontal cortex (axial view, cluster size = 191, MNI coordinates: -32, 22, -18). Greater activation found in obese than in control participants to high-calorie > low-calorie foods in (D) left amygdala (coronal view, cluster size = 33, MNI coordinates: -24, 0, -28), (E) right nucleus accumbens (coronal view, cluster size = 7, MNI coordinates: 6, 14, -8), (F) enhanced view of E. Conventions as in Fig. 4.

Group comparisons: low-calorie food condition. Results were mixed for the low-calorie foods vs. cars contrast (Table 3). The obese group showed greater activation than controls in the caudate and the putamen. Controls showed
greater activation than the obese group in right ACC and Med PFC and bilaterally in Lat OFC.

*Group comparisons: differences in activation to high-calorie and low-calorie foods.* The rationale for testing the differences between the activation in response to the high-calorie foods minus the low-calorie foods is that this comparison should most clearly reflect the response to relative reward value, with effects related to other food-associated factors subtracted out. Table 4 lists the ten ROI’s in which the contrast of high-calorie vs. low-calorie foods was significantly greater in the obese group than in controls, and Table 5 shows mean percentage activation for those ROI’s (e.g., Fig. 5D, AMYG; Fig. 5E & 5F, NAc). It is noteworthy that these effects were mostly left lateralized. The high-calorie vs. low calorie difference was greater in controls in one region, right dorsal caudate. Controls also showed greater activation than obese for the low-calorie vs. high-calorie contrast in right Lat OFC. There were no regions where obese subjects were greater than controls for this contrast.

In line with our observation that sweet high-calorie foods produced greater activation than savory foods in ACC, insula, HIPPO, caudate, and putamen in the obese group, we also found that the sweet vs. savory difference was larger in obese subjects than in controls in the same ROI’s (Supplementary Table 2). Controls showed a greater sweet vs. savory difference than the obese group in Med PFC.
Table 4.

Between-group (obese vs. control group) comparisons contrasting differences in the high- vs. low-calorie food conditions.

<table>
<thead>
<tr>
<th>Contrast</th>
<th>ROI</th>
<th>Hem</th>
<th>Cluster</th>
<th>BA</th>
<th>t</th>
<th>MNI coordinates</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>OB &gt; CTRL: HiCal &gt; LoCal</strong></td>
<td>Med OFC</td>
<td>L</td>
<td>9</td>
<td></td>
<td>2.90</td>
<td>-22, 14, -14</td>
</tr>
<tr>
<td></td>
<td>Lat OFC</td>
<td>L</td>
<td>65</td>
<td></td>
<td>3.69</td>
<td>-32, 28, -12</td>
</tr>
<tr>
<td></td>
<td>Med PFC</td>
<td>L</td>
<td>42</td>
<td>8</td>
<td>4.87</td>
<td>-4, 26, 44</td>
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<tr>
<td></td>
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<td>26</td>
<td>32</td>
<td>3.13</td>
<td>-10, 24, 26</td>
</tr>
<tr>
<td></td>
<td>Insula</td>
<td>L</td>
<td>11</td>
<td></td>
<td>3.18</td>
<td>-38, -16, -8</td>
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<td></td>
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<td>R</td>
<td>7</td>
<td></td>
<td>3.75</td>
<td>6, 14, -8</td>
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<tr>
<td></td>
<td>AMYG</td>
<td>L</td>
<td>33</td>
<td></td>
<td>5.38</td>
<td>-24, 0, -28</td>
</tr>
<tr>
<td></td>
<td>Vent Pall</td>
<td>L</td>
<td>7</td>
<td></td>
<td>2.89</td>
<td>-24, -8, 0</td>
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<td></td>
<td>HIPPO</td>
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<td>2.95</td>
<td>-20, -36, 2</td>
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<td></td>
<td>Caudate</td>
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<td>10</td>
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<td>3.13</td>
<td>-8, 18, -6</td>
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<tr>
<td><strong>CTRL &gt; OB: HiCal &gt; LoCal</strong></td>
<td>Caudate</td>
<td>R</td>
<td>15</td>
<td></td>
<td>3.22</td>
<td>22, 24, 12</td>
</tr>
<tr>
<td><strong>CTRL &gt; OB: LoCal &gt; HiCal</strong></td>
<td>Lat OFC</td>
<td>R</td>
<td>76</td>
<td></td>
<td>4.40</td>
<td>44, 28, -8</td>
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There were no significant activations for OB > CTRL, LoCal > HiCal. Conventions as in Tables 1 and 3.
Table 5.

Mean percentage signal change for the high-calorie and low-calorie food conditions for the obese and control groups

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th></th>
<th>Obese</th>
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<tr>
<td></td>
<td>HC</td>
<td>LC</td>
<td>HC</td>
<td>LC</td>
</tr>
<tr>
<td>Med OFC</td>
<td>-0.020</td>
<td>0.091</td>
<td>0.241</td>
<td>0.056</td>
</tr>
<tr>
<td>Lat OFC</td>
<td>-0.088</td>
<td>0.101</td>
<td>0.267</td>
<td>0.099</td>
</tr>
<tr>
<td>Med PFC</td>
<td>0.007</td>
<td>0.147</td>
<td>0.223</td>
<td>0.138</td>
</tr>
<tr>
<td>ACC</td>
<td>-0.035</td>
<td>0.088</td>
<td>0.194</td>
<td>0.015</td>
</tr>
<tr>
<td>Insula</td>
<td>-0.125</td>
<td>-0.056</td>
<td>0.102</td>
<td>-0.035</td>
</tr>
<tr>
<td>NAc</td>
<td>-0.110</td>
<td>-0.030</td>
<td>0.026</td>
<td>-0.116</td>
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<td>AMYG</td>
<td>0.066</td>
<td>0.132</td>
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<td>0.192</td>
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<td>Vent Pall</td>
<td>0.065</td>
<td>0.082</td>
<td>0.296</td>
<td>0.157</td>
</tr>
<tr>
<td>HIPPO (L)</td>
<td>0.194</td>
<td>0.238</td>
<td>0.323</td>
<td>0.116</td>
</tr>
<tr>
<td>HIPPO (R)</td>
<td>0.303</td>
<td>0.438</td>
<td>0.553</td>
<td>0.424</td>
</tr>
<tr>
<td>Caudate</td>
<td>-0.014</td>
<td>0.124</td>
<td>0.198</td>
<td>0.038</td>
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</tbody>
</table>

The mean percent signal change was found for the voxel of maximum activation in ROIs for which the contrast of high-calorie vs. low-calorie foods was significantly greater in the obese group compared to the control group as calculated by MarsBar (Brett et al., 2002; see Table 4). Conventions as in Table 1. HC, high-calorie; LC, low-calorie; L, left; R, right

Discussion

Our neuroimaging data show the predicted increase in reactivity to foods, high-calorie foods in particular, in obese subjects within brain regions thought to mediate motivational and emotional responses to foods and food cues (Berthoud, 2004, 2006). These findings are consistent with psychological literature indicating that food cues have increased motivational properties in obese individuals (Mela, 2006) and suggest a possible neural substrate for this effect.
**Comparisons of obese and control groups**

We found that obese women showed a greater difference between high-calorie food pictures and control images than normal-weight controls did in Med and Lat OFC, Med PFC, ACC, insula, AMYG, NAc, ventral pallidum, HIPPO, caudate, and putamen. When the groups were compared with regard to the difference in activation elicited by high- vs. low-calorie foods, results were similar except for (1) the lack of a significant difference in the putamen, and (2) a generally left lateralized pattern of activation for high- vs. low-calorie foods which was not apparent when high-calorie foods and cars were contrasted. These results with high-calorie foods contrast sharply with group comparisons of effects of low-calorie foods compared to control stimuli, for which there was no general pattern of one group of participants responding more strongly than the other.

Previous neuroimaging studies have reported obese vs. control differences in responses to visual food cues, although observed differences were not as extensive as in the present study. A PET study of regional cerebral blood flow of obese and normal-weight women during exposure to food (a lunch placed in front of the participant) revealed evidence of greater responsiveness in the parietal and temporal cortices of the obese women (Karhunen et al., 1997). Most relevant to our study, Rothemund et al. (2007) found that obese individuals showed increased activation in the dorsal striatum (caudate and putamen), anterior insula, hippocampus, and parietal lobule compared to normal-weight controls for the contrast of high-calorie foods vs. control stimuli. For high-calorie foods vs. low-calorie foods, the only group difference was in the putamen. The
reasons we found more extensive group differences than these previous studies are probably due to methodological differences in our study. For example, we tested hungry (8-9 h fasted) participants rather than participants described as neither hungry nor satiated as in Rothemund et al. (2007). In addition, our comparisons probably had greater statistical power because we imaged more time points from each subject, because we used all novel images to minimize habituation, and because of better matching of the obese and control groups to minimize variability. In particular, we controlled for menstrual cycle phase in light of evidence that reward- and emotion-related brain activation varies across the menstrual cycle. Dreher and colleagues (2007) found that monetary reward delivery resulted in greater BOLD activation during the follicular than the luteal phase of the menstrual cycle in the amygdala, caudate, midbrain, inferior frontal gyrus, and fronto-polar cortex, whereas during the luteal vs. the follicular phase, there was greater activation in the intraparietal and inferior temporal cortices. In addition, estradiol level, which rises from low to peak levels during the follicular phase and then falls again during the luteal phase, may affect cerebral perfusion (Dietrich et al., 2001).

Our fMRI data strongly parallel results from behavioral studies which have suggested that exaggerated reactivity to food cues may be an etiologic factor in obesity. Obese individuals have been found to be hyperresponsive to food cues in a wide variety of assessments (e.g., Braet and Crombez, 2003; Halford et al., 2004). Results such as increased salivation in response to food cues suggest that these stimuli have greater incentive value in the obese (Epstein et al., 1996;
Johnson and Wildman, 1983). Jansen and colleagues (2003) reported that the taste or smell of snack foods triggered overconsumption by overweight children relative to those of normal weight on a subsequent intake test. Johnson (1974) found that instrumental responding by obese participants, but not by controls, was enhanced if the reward, a preferred sandwich, was prominently displayed. Tetley et al. (2006) reported that magnitude of appetitive effects of the sight or smell of food was positively correlated with BMI. Most of these studies have used foods of high palatability and caloric value as the stimuli. In conjunction with data showing that obese individuals show stronger preferences for such foods than do normal-weight individuals (Drewnowski et al., 1985, 1992; Le Noury et al., 2002; Mela, 2001; Mela and Sanchetti, 1991; Rissanen et al., 2002), our findings support the hypothesis that exaggerated food-cue reactivity in the obese is specific, or at least most pronounced, for high-calorie foods.

Control group activations.

Comparisons of activation elicited by food images compared to non-food images for our control participants revealed results generally similar to those of previous neuroimaging studies, which have found activation in response to visual food-related stimuli in similar reward and associated areas (Arana et al., 2003; Beaver et al., 2006; Del Parigi et al., 2002; Hinton et al., 2004; Holsen et al., 2005; Killgore et al., 2003; Killgore and Yurgelun-Todd, 2005; Kinomura et al., 1994; Kringlebach et al., 2003; LaBar et al., 2001; O’Doherty et al., 2002; Porubska et al., 2006; Rothemund et al., 2007; Simmons et al., 2005; Small et
Significant activation by at least one food category was found in all of our ROI’s but the only activated subcortical ROI was HIPPO. Although activation was modulated by food category in several ROI’s, the only one showing greater activation by high- compared to low-calorie foods was the dorsal caudate. Low-calorie foods elicited greater activation than high-calorie foods in three regions of frontal cortex, Lat OFC, Med PFC, and ACC. Valence ratings from the control group did not parallel these differences because ratings of high- and low-calorie foods were not significantly different. It has been suggested that valence ratings are relatively poor measures of the incentive value of foods compared to a behavioral assessment of reinforcement value (Saelens & Epstein, 1996). It would be of interest to determine whether differences in activation of our ROI’s by high- and low-calorie food images are better paralleled by differences in reinforcement value of such foods than by valence.

Obese group activations

In obese participants, high-calorie food images elicited more activation than control stimuli in all of our ROI’s but HYPO. Although low-calorie foods were generally effective, they did not produce significant activation in Med OFC, VTA, ventral pallidum, or HYPO. High-calorie foods were significantly more effective than low-calorie foods in Lat OFC, Med PFC, AMYG, ventral pallidum, HIPPO, and putamen. No ROI’s were found in which low-calorie foods produced more activation than high-calorie foods. Thus, it is noteworthy that in those ROI’s in
which activation was modulated by food category, this effect was generally in the opposite direction in the obese and control groups. This difference, extending our conclusions based on the between-groups analyses, suggests that differences in reactivity to food pictures between obese and non-obese individuals are not merely quantitative, because different types of foods provoke the strongest activation in the two groups.

Regression analysis predicting the magnitude of the high- vs. low-calorie contrast in the obese group revealed significant positive correlations in right Med OFC, Med PFC, and putamen. None of these was a region in which the high-calorie vs. low calorie difference was significantly greater in the obese group than in controls, but they represent additional areas in which responsiveness to high-calorie foods is greater in individuals with higher BMI’s. It may be noteworthy that the right Med OFC activation was homotopic with the left Med OFC focus in which the high- vs. low-calorie contrast was greater in the obese than in controls.

When sweet and savory high-calorie foods were compared, pictures of sweet foods produced stronger activation than savory foods did bilaterally within the insula, HIPPO, caudate, and putamen and within the right ACC. The functional significance of differences in these regions rather than those thought to relate more directly to reward value, such as OFC and NAc, is not clear. However, it is interesting that sweet high-fat foods such as doughnuts, ice cream, and cake are prominent among the most preferred foods listed by obese women, but not men, and that the strength of this preference may correlate positively with BMI (Drewnowski & Holden-Wiltse, 1992; Drewnowski et al., 1992). Likewise,
among women consumption of sweet high-fat foods has been reported to correlate positively with BMI (Macdiarmid et al., 1998).

Some previous studies have compared activation elicited by sets of high- and low-calorie foods similar to ours. When these contrasts were performed, Rothemund et al. (2007) reported no significant areas of differential activation in either their obese or control groups. A study by Killgore and colleagues (2003) with normal-weight participants identified a number of areas responding differently to images of high- and low-calorie foods, but their results were generally mixed with regard to which food category was more effective. However, similar to our observations, they found increased activation to low- compared to high-calorie foods in Med OFC and that the AMYG was not differentially responsive to the two categories. Finally, using categories labeled “appetizing” and “bland” which may be approximately equivalent to our high- and low-calorie foods, respectively, Beaver et al. (2006) found greater activation to the appetizing pictures in a large number of areas. These included two of our ROI’s, ventral striatum and insula, although we did not find that their activity was modulated by food category in our control group. Thus, the weight of available evidence suggests that numerous areas respond differently according to the apparent caloric content of pictured foods, but there is, as yet, no consensus concerning the specific areas involved and the critical factors governing this modulation.
Similar substrates for obesity and drug addiction?

Our results are consistent with the hypothesis that the same network of structures showing exaggerated responsiveness to drug cues in addiction is also hyperreactive to visual food cues in obese individuals. Models hypothesizing common substrates for drug addiction and obesity implicate a hyperactive reward system involving the mesocorticolimbic dopamine system (e.g., Volkow and Wise, 2005) as well as abnormalities in regions mediating learning and memory, inhibitory control, and decision-making (Baler and Volkow, 2006; Volkow and O’Brien, 2007). As discussed previously, this and other neuroimaging studies have shown responses to food cues in many of the same regions, suggesting roles in mediating motivational effects of such cues. For example, the OFC shows multimodal responses to food cues and has been proposed to encode reward value or valence of the associated foods (Kringelbach, 2005; Rolls, 2004; 2005; 2007). In an fMRI study with human participants, both the OFC and AMYG were shown to develop responses to visual stimuli paired with subsequent oral delivery of glucose in a Pavlovian conditioning paradigm (O’Doherty et al., 2002). In animal studies, the AMYG and a prefrontal region including the OFC have been implicated in conditioned potentiation of feeding (Petrovich & Gallagher, 2007; Petrovich et al., 2005). Thus, these regions appear to be critical substrates mediating the acquisition of motivational properties by stimuli associated with food (e.g., Holland & Petrovich, 2005). Moreover, repeated stimulation of the basolateral amygdala in rats has been reported to produce hyperphagia and excessive weight gain (Loscher et al., 2003). It is also worth noting that clinical
studies have revealed that dysfunction in the OFC is associated with compulsive behaviors related to both eating and addiction (Baler & Volkow, 2006; Volkow & Fowler, 2000; Whitwell et al., 2007). It is likely, therefore, that elevated responses in the obese, especially in response to pictures of high-calorie foods such as we have demonstrated here in the AMYG and OFC, contribute to the exaggerated psychological reactivity to food cues exhibited by such individuals and may predispose them to overeating.

The NAc may also represent the reward value of food and has been conceived as an interface between areas mediating reward and the energy balance mechanisms of the hypothalamus (Kelly, 2004; O’Doherty et al., 2006). In a circuit involving reciprocal connections with the ventral pallidum, the NAc is involved in the mediation of emotional and motivational responses to food (Smith & Berridge, 2007). Furthermore, in animal studies pharmacological manipulations of the NAc have been found to stimulate food intake (Kelly, 2004; Smith & Berridge, 2007). Effects of NAc manipulations on food intake may be mediated by activation of orexin- and neuropeptide Y-containing neurons and deactivation of proopiomelanocortin-containing neurons within the hypothalamus (Zheng et al., 2003). Thus, via the NAc, excessive activation of the reward system in obese individuals could bias hypothalamic regulatory mechanisms in such a way that appetite is increased and satiety is decreased.

The ROI’s that we have referred to as associated areas appear to have motivationally relevant functions and to have important interactions with the reward system proper. The observed enhancement of responsiveness in any or
all of these regions could contribute to exaggerated appetitive motivation in obese individuals in response to food cues. The (rostral) insula, for example, is the location of primary taste cortex (Rolls, 2007), and the insula is also involved in motivational processes (Naqvi et al., 2007; Porubska et al., 2006). The dorsal striatum appears to process rewarding action-outcomes and has been associated with habit learning and craving in addiction (O'Doherty et al., 2004; Volkow et al., 2006). There is evidence that Med PFC and rostral ACC process the relative value of an anticipated reward and use that information in mediating action selection between competing responses (Amodio & Frith, 2006; Marsh et al., 2006). The hippocampus has been implicated in incentive motivation, an action which may involve its modulation of dopamine release in the NAc (Louilot & Moal, 1994; Tracy et al., 2001). Activation in HIPPO has been associated with food and drug craving and with viewing of reward-predicting pictures (Pelchat et al., 2004; Wang et al., 2007; Wittman et al., 2005). It is noteworthy that the greatest percentage signal change values that we observed in both groups were in the right HIPPO, possibly reflecting activation of memories associated with the pictured foods, although there were not dramatic differences in activation produced by high- and low-calorie foods.

As has been proposed with regard to addiction, abnormalities in regions involved in inhibitory control may also contribute to obesity. Recently, Alonso-Alonso and Pascual-Leone (2007) proposed that hypoactivation in the right prefrontal cortex of obese individuals is related to poor cognitive control of food intake. In our study, we found less activation in a region in the right Med PFC in
the obese group relative to the controls for the high-calorie foods vs. cars contrast. Other studies have suggested that cognitive or regulatory control of food intake may be mediated by dorsolateral prefrontal cortex (Del Parigi et al., 2007; Le et al., 2006; Uher et al., 2004; 2005). Further research is needed to determine whether the area of Med PFC in which we found hypoactivation is also involved in cognitive dysfunction related to obesity.

**Caveats**

Although we observed clear and extensive differences between our obese and control samples, because of our cross-sectional experimental design, it is not possible to determine whether the observed effects represent causes or consequences of obesity. Also, we studied a rather homogeneous group of obese and control subjects: all young-adult women with BMI’s between 30-41 or 20–25, respectively, without an eating disorder, fasted for 8-9 hours, in the follicular stage of the menstrual cycle. These similarities probably increased our power by reducing spurious variability, but further research is needed to assess the generality of our findings. For example, it would be interesting to compare results obtained with subjects in fasted and satiated states.

With the imaging parameters that we used, we saw signal dropout in the medial OFC. Because of this, we were not able to fully evaluate potential within- and between-group differences in this region. We do not, however, believe that this consideration invalidates differences that were observed.
Summary and conclusions

In summary, our neuroimaging study found greater activation to high-calorie foods in obese individuals compared to controls within a wide range of brain regions thought to mediate motivational and emotional responses to foods and food cues. These results strongly support a hypothesis based on behavioral studies which proposes that overeating in obese individuals is triggered by exaggerated reactivity to stimuli associated with high-calorie foods. Functional implications of the current results could be addressed in longitudinal studies. It might be expected, for example, the exaggerated reward system activation to high-incentive food cues would predict weight gain, especially in subjects at risk for obesity. Conversely, magnitude of activation to high-incentive food cues might discriminate between those who were subsequently successful or unsuccessful in losing weight and/or maintaining weight loss. It would be of interest to determine whether the responsiveness of a hyperactive reward system could be moderated in response to successful anti-obesity therapy and whether such neuroadaptation would lead or follow the weight loss.
Acknowledgments

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References


Supplementary Material

Supplementary Table 1.

Within-group (obese) BMI regression results for the high- > low-calorie food conditions contrast.

<table>
<thead>
<tr>
<th>ROI</th>
<th>Hem</th>
<th>Cluster</th>
<th>BA</th>
<th>r</th>
<th>t</th>
<th>MNI coordinates</th>
</tr>
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<tbody>
<tr>
<td>Med OFC</td>
<td>R</td>
<td>7</td>
<td>7</td>
<td>.73</td>
<td>3.42</td>
<td>20, 18, -14</td>
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<tr>
<td>Med PFC</td>
<td>R</td>
<td>25</td>
<td>25</td>
<td>.86</td>
<td>5.33</td>
<td>12, 26, 52</td>
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<tr>
<td>Putamen</td>
<td>R</td>
<td>13</td>
<td>13</td>
<td>.78</td>
<td>3.92</td>
<td>18, 12, -10</td>
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</tbody>
</table>

r = correlation coefficient for the relationship between BMI and ROIs. Other conventions as in Table 1.
Supplementary Table 2.

Between-group (obese vs. control group) comparisons contrasting differences in the sweet vs. savory food conditions.

<table>
<thead>
<tr>
<th>Contrast</th>
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<tr>
<td><strong>OB &gt; CTRL:</strong></td>
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<tr>
<td>Sweet &gt; Savory</td>
<td>ACC</td>
<td>R</td>
<td>7</td>
<td>32</td>
<td>3.03</td>
<td>18, 44, 8</td>
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<td></td>
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<tr>
<td>Sweet &gt; Savory</td>
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<td><strong>OB &gt; CTRL:</strong></td>
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<tr>
<td>Savory &gt; Sweet</td>
<td>ACC</td>
<td>R</td>
<td>8</td>
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<tr>
<td></td>
<td>Caudate</td>
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<td>8</td>
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Conventions as in Tables 1 and 3.
INTEROCEPTIVE AND EXTEROCEPTIVE FACTORS PREDICT DIFFERENT PATTERNS OF REWARD-SYSTEM ACTIVATION IN RESPONSE TO FOOD IMAGES IN OBESE AND NORMAL-WEIGHT INDIVIDUALS

by

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In preparation for Appetite
Format adapted for dissertation
Abstract

Studies have indicated that obese individuals are less sensitive to interoceptive cues like hunger and rely more on exteroceptive cues such as the sight of foods to determine ingestive behavior. Food cues, especially those associated with high-calorie foods, also appear to stimulate relatively more non-homeostatic eating in obese individuals. The neural underpinnings of these contributors to obesity might be overreactive reward system brain regions. We investigated reward-related brain activation in response to pictures of high-calorie and low-calorie foods predicted by (1) hunger and (2) the hedonic value and incentive salience of the food stimuli in 12 obese compared to 12 normal-weight women using fMRI. For the hunger analyses, the obese group had relatively less hunger-modulated reward system activation in response to food images, especially low-calorie food images. We also found that the obese individuals had relatively greater reward system activation predicted by foods’ incentive salience more so than by their hedonic value, especially for high-calorie foods. Heightened incentive salience of food cues, an exteroceptive factor, coupled with a deficient modulatory effect of hunger may promote an increased drive for foods and food cues, hyperphagia, and subsequent weight gain in obese individuals.

Keywords: interoceptive, exteroceptive, hunger, liking, wanting, obesity, reward
The initiation of food intake is influenced by the interaction of interoceptive factors related to energy depletion giving rise to hunger and exteroceptive factors from the environment such as the sight or smell of foods (Cornell et al., 1989; Marcelino et al., 2001; Tuomisto et al., 1998). Compared to normal-weight individuals, obese individuals appear to rely less on interoceptive cues and more on exteroceptive factors to determine meal initiation (Doucet et al., 2003; Hetherington et al., 1996; Schachter, 1968; Tuomisto et al., 1998; Vincent and Le Roux, 2008; Wansink et al., 2007). Stimuli associated with energy dense, nutrient poor foods high in fat can stimulate non-homeostatic eating more in obese than normal-weight individuals and might be an important contributing factor to obesity (Berthoud, 2004; Mela, 2006; Saelens & Epstein, 1996; WHO, 2003). The neural substrate for this effect appears to be exaggerated reactivity of the reward system. Recently, we found widespread brain activation differences in fasted obese compared to normal-weight women in response to pictures of high-calorie foods in areas that have been associated with reward functioning, including medial (Med) and lateral (Lat) orbitofrontal cortex (OFC), Med prefrontal cortex (PFC), anterior cingulate cortex (ACC), insula, amygdala (AMYG), nucleus accumbens (NAc), ventral pallidum, hippocampus (HIPPO), caudate, and putamen (Stoeckel et al., 2008). Other neuroimaging studies have reported similar, but less extensive, differences related to body mass (Del Parigi et al., 2002, 2004, 2005; Gautier et al., 2000; James et al., 2004; Killgore and
Although obesity may be explained, in part, by differences in interoceptive and exteroceptive factors, there have been no neuroimaging studies investigating how both of these factors might modulate reward-related brain activation, especially in the same individuals. There is also strong evidence that reward can be parsed into separate neurobiological and psychological components related to hedonic vs. motivational processes, and this distinction may have important implications for obesity (e.g., Berridge, 1996; Finlayson et al., 2007). Berridge (1996) proposed a terminology to refer to the affective and motivational components of reward dubbed “liking” and “wanting”, respectively. Incentive salience or “wanting” is the non-homeostatic desire to consume food, whereas hedonics or “liking” is conceptualized as an affective appraisal of pleasure related to foods or food cues. Tasks involving instrumental performance have shown that the reinforcement value of food (a measure of “wanting”) was greater for obese compared to lean individuals (e.g., Epstein and Leddy, 2006; Johnson, 1974; Saelens & Epstein, 1996). By contrast, a number of studies have failed to find that obese individuals rated high-calorie foods as more pleasant (a measure of “liking”) than did those of normal weight (e.g., Cox et al., 1998; Stoeckel et al., 2007). The “wanting” component of the reward system is likely regulated by the mesolimbic dopamine system including ventral tegmental area (VTA) and AMYG (e.g., Berridge and Robinson, 2003). “Liking” is likely mediated by the opioid system, particularly in the NAc and ventral pallidum (e.g., Berridge and
Robinson, 2003; Smith and Berridge, 2005). Some areas, e.g., NAc, insula, and OFC, have been reported to function in both “liking” and “wanting.”

There have been few neuroimaging studies investigating selective brain activation in response to the hedonic value or incentive salience of food cues. These studies have reported increased activation in OFC, ACC, and insula for “liking,” and in OFC, insula, and dorsal striatum (i.e., putamen) for “wanting” (De Araujo et al., 2003a, 2003b; Kringelbach, 2004, 2005; Porubska et al., 2006). To our knowledge, the current study is the first human functional neuroimaging study to investigate both “liking” and “wanting” in response to food cues in the same obese and normal-weight participants.

Whereas “liking” and “wanting” mechanisms involve the external appraisal of foods and often serve non-homeostatic functions, hunger is usually conceptualized as relating to the interpretation of the internal milieu with regard to homeostatic state (e.g., Finlayson et al., 2007). The hedonic value of a food is modulated by motivational state, with food perceived as pleasant during a state of hunger and unpleasant or neutral during satiety, a phenomenon termed “alliesthesia” (Cabanac, 1971; Fantino, 1984). Alliesthesia appears to function differently in obese compared to healthy-weight individuals. For example, Blundell & Hill (1988) found that glucose-induced negative gustative alliesthesia (i.e., the taste of glucose becomes less pleasant with increased intake) was present in lean, but not in obese participants. This may indicate that compared to normal-weight individuals, obese individuals are not as sensitive to interoceptive
cues that serve to modulate hedonics and subsequent food intake as a function of homeostatic need.

Neuroimaging studies have found activation related to fasting state in normal-weight individuals in the insula, OFC, ACC, HIPPO, caudate, and putamen (DelParigi et al., 2002; Tataranni et al., 1999). Compared to normal-weight controls, fasted obese individuals had greater brain activation in insula, HIPPO, and OFC (DelParigi et al., 2005). Hunger has been shown to modulate response to food images in normal-weight participants in insula, OFC, ACC, and AMYG (Fuhrer et al., 2008; LaBar et al., 2001; Morris and Dolan, 2001; Uher et al., 2006; Wang et al., 2004). There have been no neuroimaging studies looking at differences in obese and normal-weight individuals as a function of hunger in response to food stimuli, especially visual food stimuli. The current study is also the first to investigate how subjective hunger modulates reward-related brain activation, specifically in response to foods in obese compared to normal-weight participants.

The purpose of the present study is to measure how subjective interoceptive (hunger) and exteroceptive (hedonic value and incentive salience) factors modulate brain activation in the reward system in response to visual food stimuli in obese and normal-weight women so that we may better understand the mechanisms that lead to obesity. We hypothesize that subjective pleasantness ratings (“liking”) and appetite ratings (“wanting”) of food images will predict different patterns of brain activation coding hedonic value (“liking”) and incentive salience (“wanting) in response to food images in reward-related areas. Obese
participants will also show greater regional differences compared to normal-weight controls in “wanting” rather than “liking” areas. In addition, we hypothesize that subjective hunger will modulate reward-related brain activation in response to foods more in the normal-weight than the obese group.

**Materials and methods**

*Participants*

Participants were 12 obese (Body Mass Index, BMI, = 30.8 – 41.2) and 12 normal-weight (BMI = 19.7 – 24.5) right-handed women recruited from the University of Alabama at Birmingham community. There were no group differences on mean age (obese: 27.8, SD = 6.2; control: 28.0, SD = 4.4), ethnicity (obese: 7 African-American, 5 Caucasian; control: 6 African-American, 6 Caucasian), education (obese: 16.7 years, SD = 2.2; control: 17.2, SD = 2.8), or mean day of the menstrual cycle (all in follicular phase, obese: day 6.8, SD = 3.1, control: day 5.7, SD = 3.3). Individuals were excluded based on criteria including a positive eating disorder history, active dieting or participating in a weight-loss program, or weight > 305 pounds (138 kg) with girth > 64 inches (163 cm) due to scanner limitations. For a more detailed description, see Stoeckel et al. (2008).

*Stimuli*

The stimuli used during the imaging session consisted of 252 color pictures from a previous study (Stoeckel et al., 2008), all of consistent size, resolution, and luminance (see Stoeckel et al., 2007, for further details). The 168
food images were subdivided into low-calorie and high-calorie categories, each consisting of 84 unique images. Low-calorie food images consisted of such low-fat items as steamed vegetables and broiled fish. High-calorie foods were primarily items high in fat such as cheesecake or pizza. Control stimuli consisted of car images of a wide range of makes, models, ages, and colors. The stimuli participants rated on valence and appetite value following the imaging session included 33 foods (17 low-calorie foods and 16 high-calorie foods) and 17 cars (valence ratings only) randomly selected from the imaging session.

Response Measures

Participants provided subjective hunger ratings using three visual analog scales. The scales consisted of 150 mm lines with the left ends anchored at “none at all” and the right at “extremely” or “most possible” (Nolan et al., 2003; Stoeckel et al., 2008). Hunger was assessed using the following three questions: 1) How hungry are you? 2) How strong is your desire to eat your favorite food now? 3) How much food would you like to eat right now? Participants also rated visual stimuli on emotional valence (i.e., pleasantness; Osgood, Suci, & Tannenbaum; Russell, 1979) and subjective appetite value (Porubska et al., 2006). Valence and appetite ratings for each image were manually recorded on 9-point Likert scales. For the valence scale, a rating of 1 indicated an extremely unpleasant response to the visual image and a rating of 9 indicated an extremely pleasant response to the visual image. For the appetite scale, a rating of 1 indicated, “This food is not appetizing; I would strongly not like to eat this food
now” and 9 indicated, “This food is very appetizing; I strongly would like to eat this food now”. A rating of 5 indicated a neutral response for both scales.

Procedure

After thorough screening to validate BMI, eating disorder diagnosis, and other study criteria, participants were scheduled for the fMRI session. They were instructed to eat a normal breakfast between 7-8 A.M. but to skip lunch and consume only water so that they had fasted for approximately 8-9 h before being imaged between 3-5 P.M. Participants were asked to provide subjective hunger ratings immediately prior to the imaging session.

While participants were in the magnet, visual stimuli were presented in a block design format, with a total of six 3:09 min runs per imaging session. Each run consisted of two 21 s epochs each of cars (C), low-calorie foods (LC), and one epoch each of high-calorie foods (HC) pseudorandomly presented to the participants. Within each 21 s epoch of food or car images, seven individual images were presented for 2.5 s each followed by a 0.5 s gap each, separated by 9 s of a gray blank screen with a fixation cross. Each run consisted of 63 volumes for a total of 378 volumes across six runs, of which 84 volumes were acquired during each of the car, low-calorie food, and high-calorie food exposures. The visual images were presented by a laptop using VPM software (Cook et al., 1987). Images were projected onto a screen behind the participant’s head and viewed via a 45° single-surface rear-projecting mirror attached to the head coil. Participants were financially compensated for their participation. All
procedures were reviewed and approved by UAB’s Institutional Review Board for Human Use. For additional details, see Stoeckel et al. (2008).

**MRI acquisition and processing**

Functional MRI data were acquired using a Philips Intera 3T ultra-short bore magnet equipped with a SENSE head coil. Images were collected using a single-shot T2*-weighted gradient-echo EPI pulse sequence. We used TE = 30 msec, TR = 3 sec, and an 85º flip angle for 30 axial slices 4 mm thick with a 1 mm interslice gap, a scan resolution of 80 x 79, reconstructed to 128 x 128, and with a 230 x 149 x 230 mm FOV. The first four scans were discarded to allow the magnet to achieve steady-state magnetization.

Data were preprocessed (motion correction, normalization to the MNI coordinate system using the SPM2 EPI template, and smoothed with a 6 mm FWHM Gaussian filter) using the SPM2 software package (Wellcome Dept. Imaging Neuroscience, London, UK). No data sets failed to meet the movement inclusionary criteria, which were that movement before correction was < 2 mm in translational movement and < 2º in rotational movement. Refer to Stoeckel et al. (2008) for further details.

**Data analysis**

**Behavioral measures.** Ratings on the three hunger questions were highly correlated (Pearson’s r = .49 - .70). A principal components analysis (PCA) was used to reduce the data from the three hunger questions to one hunger score for
each participant representing the maximum total variance explained from the linear combination of the three hunger questions (SPSS 11.5, SPSS Inc., Chicago, USA). An independent samples t-test was used to compare the groups on the PCA hunger score. Valence and appetite ratings of high- and low-calorie food images were analyzed with a mixed between-group (BMI group), within-group (Food Category) analysis of variance (ANOVA). Food categories were compared within each group using simple-effects analyses (Howell, 2007).

**fMRI data.** Block-design blood oxygen level dependent (BOLD) responses were analyzed within the context of the General Linear Model on a voxel by voxel basis as implemented in SPM2 (Friston et al., 1995). The time course of brain activation was modeled with a boxcar function convolved with the canonical hemodynamic response function (HRF) and a temporal derivative function. The data were high-pass filtered (1/128 Hz) to remove low frequency drifts. A first order autoregressive model was also implemented to correct for autocorrelations in the error term of the fMRI model.

A two-stage random-effects procedure was used for the statistical analysis to account for both within-subject and between-subject variability. First, the fMRI data from each individual participant were used to generate statistical contrasts of the parameter estimates (betas) in order to test the differences between the time points associated with the high-calorie foods > control stimuli and low-calorie foods > control stimuli. In an earlier study (Stoeckel et al., 2008), we found group differences in patterns of reward-related activation, with the obese group exhibiting greater activation to high-calorie foods and controls to low-
calorie foods. Because both categories of food stimuli produced more reward-related activation compared to control stimuli in both obese and normal-weight participants, we chose to use these contrasts to determine the effect of hunger and valence and appetite ratings within- and between-groups. The contrasts for each participant were then entered into second-level analyses for within-group and between-group comparisons. The interactions of group (obese vs. controls) X hunger PCA score in predicting activation related to the contrasts of high-calorie foods > control stimuli and low-calorie foods > control stimuli were investigated using independent multiple regression models for each contrast. Follow-up within-group regression analyses were also performed using hunger PCA scores to predict activation for the contrasts of high-calorie > control stimuli and low-calorie > control stimuli, separately.

For the valence and appetite ratings analyses, each participant’s mean valence and appetite ratings for the high-calorie food stimuli and low-calorie food stimuli were entered into separate multiple regression analyses to predict activation related to the contrasts of high-calorie foods > control stimuli and low-calorie foods > control stimuli, respectively. Entering mean valence and appetite ratings into the same model allowed us to investigate activation related to each contrast explained by each measure (valence and appetite), with activation related to shared variance between these variables partialled out. For both these statistical analyses, we used a region of interest (ROI) approach to address our a priori hypotheses (Stoeckel et al., 2008). Our areas of interest were reward areas: VTA, NAc, AMYG, ventral pallidum, and OFC. We also analyzed other,
functionally associated regions that have been found to be activated in comparable fMRI and PET studies: ACC, Med PFC, insula, hippocampus (HIPPO), hypothalamus (HYPO), caudate and putamen (see Stoeckel et al., 2008 for review). Significance for these a priori ROI's was assessed at $p < .01$ with a spatial extent of seven voxels using small volume correction (SVC).

ROI's were defined using the WFU Pickatlas and the AAL and Talairach Daemon atlases (Lancaster et al., 2000; Maldjian et al., 2004; Tzourio-Mazoyer et al., 2002). Regions that were unavailable in these libraries (VTA and NAc) were drawn within the WFU Pickatlas (see Stoeckel et al., 2008 for details). MNI coordinates for the voxels of maximum activation within an ROI obtained from SPM2 were entered into the WFU Pickatlas using the AAL and Talairach Daemon atlases to determine the anatomical location of each locus. In some instances, the AAL and Talairach Daemon atlases did not identify or were inconsistent in classifying a particular locus. Therefore, the classification of activated voxels was verified by visual inspection of the data, and unidentified or misclassified loci of activation were identified using a human brain atlas (Mai et al., 2004).
**Results**

*Behavioral measures*

**Hunger ratings**

There were no group differences on the hunger PCA scores (t [22] = 0.93, p = .36). This outcome suggests that our fMRI results did not result from group differences in hunger.

**Valence ratings**

ANOVA revealed a significant group by food category interaction (F[1,22] = 4.39, p = .048). A simple effects analysis showed that obese individuals’ ratings of high calorie foods were higher than their ratings of low calorie foods (F[1,22] = 4.42, p = .047). Within the control group, ratings of high- and low-calorie foods were not significantly different (F[1,22] = 0.74, p = .40). Figure 1 shows mean valence ratings of low calorie and high calorie food images by control and obese participants.

**Appetite ratings**

The appetite rating scale was introduced into the study at the midpoint of data collection. Thus, for these analyses we have data from 5 obese and 7 normal-weight participants. Like the full study sample, there were no group differences on mean age, ethnicity, education, or mean day of the menstrual cycle. ANOVA of appetite ratings resulted in no group by food category interaction (F[1,10] = 1.74, p = .216; Fig. 1).
Fig. 1. Mean valence and appetite ratings (with SE) of food images by obese (N=12) and control (N=12) participants. *P < 0.05 for obese group’s valence ratings of high-calorie vs. low-calorie foods. OB = obese, C = controls.

fMRI results

The high-calorie food > control and low-calorie food > control contrasts X hunger rating interactions were used to determine those regions of interest where hunger ratings predicted greater brain activation related to these contrasted stimulus categories. We report ROIs showing greater brain activation related to the contrasts predicted by increased hunger ratings (i.e., positive betas or regression coefficients).
Prediction of high-calorie foods > control contrast values by hunger ratings

*Obese group.* The high-calorie food > control contrast X hunger rating interaction within the obese group indicated that increased hunger predicted greater activation to high-calorie foods in the left Lat OFC, left ACC, and right HIPPO (Table 1).

*Control group.* Controls, on the other hand, showed a positive relationship between hunger and activation to high-calorie foods in bilateral insula, bilateral AMYG, and bilateral HIPPO, with the right HIPPO cluster in the controls more anteroventral compared to the right HIPPO cluster in the obese group (Table 1).

*Obese > Control group.* There were no regions showing significantly greater regression coefficients in obese participants compared to controls.

*Control > Obese group.* Compared to obese participants, controls showed greater regression coefficients, indicating increased activation predicted by greater hunger ratings, in bilateral HIPPO (Table 1; Fig. 2a – left HIPPO). The relationship between activation and hunger resulted in a positive beta for the controls in bilateral HIPPO, meaning increased activation was predicted by increased hunger, whereas the same analysis within the obese group resulted in a negative beta in left HIPPO, meaning increased hunger led to lesser activation in this region, and a positive beta in right HIPPO.
Table 1. Regression analyses comparing groups using the hunger PCA score to predict contrast estimates from the high-calorie foods > control contrast.

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For all contrasts (high-calorie foods vs. control and low-calorie foods vs. control), activated regions of interest (ROI) as identified by the WFU PickAtlas, right or left (R, L) hemisphere (Hem), number of contiguous voxels (Cluster), approximate Brodmann areas (BA) as identified in the WFU PickAtlas*, t-values, and coordinates of the voxel of greatest activation within the Montreal Neurological Institute (MNI) coordinate system are displayed. Other abbreviations as in the text.

* Brodmann's areas (BA) were identified in the WFU PickAtlas based on their anatomical location in Talairach space after converting the MNI coordinates to Talairach coordinates. As the locations of BA as defined in the Talairach Daemon are estimates based on an approximate conversion, the BA of some of the cortical foci in our study remained unidentified by the WFU PickAtlas.
Prediction of low-calorie foods > control contrast values by hunger ratings

**Obese group.** The low-calorie food > control contrast X hunger rating interaction within the obese group indicated increased hunger predicted greater activation to the low-calorie food stimuli in only one region (VTA, Table 2).

**Control group.** Compared to the obese group, controls showed many more ROIs in which greater hunger predicted significantly greater activation to low-calorie foods: bilateral Lat OFC, bilateral Med PFC, bilateral ACC, bilateral insula, bilateral AMYG, left NAc, bilateral HIPPO, and right putamen (Table 2).

**Obese > Control group.** There were no regions showing significantly greater activation to low-calorie foods > control stimuli predicted by hunger ratings in obese participants compared to controls.

**Control > Obese group.** Compared to the obese group, controls had greater regression coefficients, signifying greater hunger predicted increased activation, in right Lat OFC, bilateral Med PFC, right ACC, bilateral insula, and right putamen (Table 2; Fig. 2b – left insula). The relationship between activation and hunger resulted in a positive beta for the controls and a negative beta for the obese group in all of these ROIs.
Table 2. Regression analyses comparing groups using the hunger PCA score to predict contrast estimates from the low-calorie foods > control contrast.

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Conventions as in Table 1.
Fig. 2. fMRI activation in the HIPPO (A, coronal section; graph, y-axis in arbitrary units) from the high-calorie foods > control contrast predicted by hunger PCA scores in obese and control participants. fMRI activation in the insula (B, coronal section, graph) from the low-calorie foods > control contrast predicted by hunger PCA scores in the two groups. Activation is overlaid on the SPM2 single-subject T1 template. Color bar indicates t-values.
Prediction of high-calorie foods > control contrast values by valence and appetite ratings

The valence and appetite ratings were entered into the multiple regression model simultaneously in order to partition out the unique effects of each of these variables in predicting the high-calorie foods > control images and low-calorie foods > control images contrast values. We report ROIs showing greater brain activation related to the contrasts predicted by increased (positive betas) valence and appetite ratings.

### Valence Ratings

**Obese group.** Within the obese group, there was a significant valence ratings × contrast interaction, indicating increased valence ratings predicted greater activation in response to high-calorie foods in right Lat OFC and right ventral pallidum (Table 3).

**Control group.** The normal-weight controls showed significantly greater activation predicted by increased valence ratings in bilateral Med OFC, Lat OFC, Med PFC, ACC, insula, HIPPO, NAc, and putamen and right AMYG and caudate (Table 3).

**Obese > Control group.** Compared to controls, the obese group had a greater regression coefficient for the high-calorie food > control contrast predicted by increased valence ratings in right ventral pallidum (Table 3). The
relationship between activation and valence ratings resulted in a positive beta for the obese group and a negative beta in the controls.

**Control > Obese group.** The same analysis resulted in greater activation in controls compared to the obese group in left Lat OFC, bilateral Med PFC, left ACC, bilateral insula, right caudate, and left putamen (Fig 3, Lat OFC; Table 3). As in the obese > control group analysis, the relationship between activation and valence ratings in the ROIs showing a significant control > obese difference resulted in a positive beta for the controls and negative beta in the obese group.

![fMRI activation in the Lat OFC](image)

Fig. 3. fMRI activation in the Lat OFC (axial section; graph, y-axis in arbitrary units) from the low-calorie foods > control contrast predicted by valence scores in obese and control participants. Conventions as in Fig. 2.

**Appetite Ratings**

**Obese group.** The obese participants showed greater brain activation predicted by increased appetite ratings in left Lat OFC, bilateral Med PFC, bilateral ACC, and left caudate (Table 3). These results illustrate a pattern of
more extensive activation predicted by appetite ratings compared to valence ratings in the obese group.

*Control group.* The appetite ratings X food > control contrast for the controls resulted in increased appetite ratings predicting greater contrast values for right insula only (Table 2). Compared to the results showing extensive cortical and subcortical activation predicted by valence ratings in the control group, the appetite results indicate significant activation for fewer cortical ROIs and no subcortical ROIs in response to foods.

*Obese > Control group.* Compared to normal-weight controls, the obese group had greater high-calorie food > control contrast values predicted by increased appetite ratings in bilateral Med PFC and left caudate (Table 3). The high-calorie food > control contrast X appetite ratings interaction resulted in a positive beta for the obese group and a negative beta in the controls for the ROIs showing the obese > control group difference.

*Control > Obese group.* There were no regions showing greater activation in normal-weight controls compared to the obese group predicted by appetite ratings (Table 3).

The valence ratings comparison resulted in the most extensive group differences between obese and normal-weight controls, with controls showing greater activation in numerous, mostly cortical regions compared to the obese group. The appetite ratings, on the other hand, resulted in a group difference only in the obese > control comparisons.
Table 3. Regression analyses comparing groups using the valence and appetite ratings to predict contrast estimates from the high-calorie foods > control contrast.

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<td>8.73</td>
<td>-16, 18, -6</td>
</tr>
<tr>
<td>CTRL &gt; OB (Appetite)</td>
<td>NONE</td>
<td></td>
<td></td>
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</table>

Conventions as in Table 1.
Prediction of low-calorie foods > control contrast values by valence and appetite ratings

Valence Ratings

Obese group. There were no significant valence ratings X contrast interactions, indicating increased valence ratings did not predict greater values for the low-calorie > control stimuli contrast (Table 4).

Control group. The normal-weight controls showed greater activation predicted by increased valence ratings in left insula only (Table 4).

Obese vs. Control group. There were no group differences using increased valence ratings to predict greater low-calorie food > control contrast values (Table 4).

Appetite Ratings

Obese group. The obese group showed greater brain activation predicted by increased appetite ratings in right ACC only (Table 4).

Control group. The controls, on the other hand, had greater contrast values in right Lat OFC, bilateral Med PFC, bilateral insula, and right putamen predicted by increased appetite ratings (Table 4). Compared to the valence results, there was more extensive, mostly cortical, activation predicted by appetite ratings in the control group.

Obese vs. Control group. The only group difference for greater low-calorie > control contrast values predicted by increased appetite ratings for the obese > control comparison was in a small cluster (~56 mm^3) in right ACC (Table 4).
Compared to the low-calorie foods, high-calorie foods resulted in much more activation and many more group differences in activation predicted by valence and appetite ratings.

Table 4. Regression analyses comparing groups using the valence and appetite ratings to predict contrast estimates from the low-calorie foods > control contrast.

<table>
<thead>
<tr>
<th>Contrast</th>
<th>ROI</th>
<th>Hem</th>
<th>Cluster</th>
<th>BA</th>
<th>t</th>
<th>MNI coordinates</th>
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<tbody>
<tr>
<td>Obese (Valence)</td>
<td>NONE</td>
<td></td>
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<td></td>
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<td></td>
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<tr>
<td>(Appetite)</td>
<td>ACC</td>
<td>R</td>
<td>9</td>
<td>6, 30, 22</td>
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<tr>
<td>Controls (Valence)</td>
<td>Insula</td>
<td>L</td>
<td>13</td>
<td>-46, -10, 4</td>
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<td></td>
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<tr>
<td>(Appetite)</td>
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<td>R</td>
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<td>34, 50, -12</td>
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<td></td>
<td>Med PFC</td>
<td>L</td>
<td>17</td>
<td>-8, 30, 54</td>
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<tr>
<td></td>
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<td>R</td>
<td>60 9</td>
<td>10, 52, 40</td>
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</tr>
<tr>
<td></td>
<td>Insula</td>
<td>L</td>
<td>78 13</td>
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<td></td>
<td></td>
<td>R</td>
<td>218 13</td>
<td>44, -14, 18</td>
<td></td>
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</tr>
<tr>
<td></td>
<td>Putamen</td>
<td>R</td>
<td>8</td>
<td>36, -2, -2</td>
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<td>6, 28, 18</td>
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</table>

Conventions as in Table 1.
Discussion

Our results are consistent with the idea that interoceptive cues appear to be less effective at modulating reactivity to foods, especially low-calorie foods, in hungry obese compared to normal-weight individuals. We previously reported an increase in reactivity to foods, especially high-calorie foods, in obese subjects within brain regions thought to mediate motivational and emotional responses to food cues (Stoeckel et al., 2008). However, here we found there was little modulation of this reward system activation related to subjective hunger ratings in obese individuals. We also found that the increased potency of foods in triggering greater reward system activation in obese individuals appears to be driven by these foods’ incentive salience (“wanting”) more than their hedonic value (“liking”), and that this effect was specific to high-calorie foods. Hunger (an interoceptive factor) failing to modulate activation to foods in the obese group despite greater reward and associated brain activation in response to food stimuli in obese compared to normal-weight participants is consistent with the idea that incentive salience plays a more prominent role than hunger in driving intake in obese individuals.

Hunger

Food deprivation increases the reinforcing value (i.e., “wanting”) of food, which stimulates food intake in healthy-weight individuals (e.g., Epstein et al., 2003). However, hunger may modulate the motivational value of foods, a phenomenon known as alliesthesia, differently in obese and normal-weight
individuals. Doucet et al. (2003) found no association between reported food intake and subjective report of appetite in obese participants. An earlier study by our group found a trend for subjects of low BMI to show a greater magnitude of the alliesthesia effect (Stoeckel et al., 2007). Finally, as mentioned earlier, Blundell & Hill (1988) reported that glucose-induced negative gustative alliesthesia was present in lean, but not in obese participants. These results demonstrate that obese individuals may not detect the usual interoceptive cues that serve to modulate the reward value of food cues and subsequent food intake related to homeostatic need.

Brain imaging allows researchers to study human brain functioning in vivo and has led to further understanding of the neural mechanisms mediating hunger. Food deprivation has been found to increase brain activation in the insula, OFC, ACC, HIPPO, caudate, and putamen in normal-weight participants in positron emission tomography (PET) studies (DelParigi et al., 2002; Tataranni et al., 1999). Another PET study found that obese individuals had greater hunger-related activation than normal-weight participants in insula, HIPPO, and OFC (DelParigi et al., 2005). In neuroimaging studies examining brain activation in response to food images, increased hunger in normal-weight participants was associated with greater activation in such areas as insula, OFC, ACC, and AMYG (Fuhrer et al., 2008; LaBar et al., 2001; Uher et al., 2006; Wang et al., 2004). Using PET, Wang et al. (2004) found that in food-deprived normal-weight participants, there was an overall increase in whole brain metabolism in response to the presentation of the participant’s favorite food. There was also increased
activation of the anterior insula and orbitofrontal cortices (Wang et al., 2004). Only right orbitofrontal activation was correlated with increased subjective report of hunger and desire for food (Wang et al., 2004). Finally, in obese women with binge-eating disorder, there was a positive correlation between self-reports of hunger elicited by food and blood flow in the left prefrontal cortex (Karhunen et al., 2000).

We used participants’ subjective hunger ratings to predict brain activation to foods of differing caloric density (high- and low-calorie foods) in obese and normal-weight participants. This is the first neuroimaging study looking at the effect of hunger on the modulation of brain activation in response to high-calorie and low-calorie foods independently. Our within-group analysis for both the high-calorie and low-calorie food contrasts in normal-weight participants resulted in similar findings as in previous studies, including activation in insula (e.g., Uher et al., 2006), HIPPO (e.g., Tatarrani et al., 1999), and AMYG (LaBar et al., 2001). We found additional activation in bilateral Med PFC, bilateral ACC, left NAc, and right putamen resulting from the low-calorie foods > control contrast. There were no additional regions (other than insula, HIPPO, and AMYG) activated by the high-calorie foods > control comparison within normal-weight controls. Within the obese group, activation to the high-calorie foods was positively predicted by greater hunger ratings in left Lat OFC and small clusters in left ACC (8 voxels) and right HIPPO (7 voxels). Only a small focus (7 voxels) in VTA was activated in relation to increased hunger in response to low-calorie foods in the obese group. In addition, group comparisons showed no ROIs with regression coefficients
greater for obese than controls for either the high-calorie or low-calorie foods, but
greater coefficients in control participants in bilateral HIPPO (high-calorie foods)
and right Lat OFC, bilateral Med PFC, right ACC, bilateral insula, and right
putamen (low-calorie foods). This is in contrast to the findings of Del Parigi et al.
(2005), who reported greater hunger-stimulated activation in insula and HIPPO in
obese compared to normal-weight participants. A possible explanation for these
differences is that we were measuring how hunger modulated reward-related
activation in response to foods. The differences reported by Del Parigi et al.
(2005) were the result of group comparisons during a food deprived state vs.
sated state following a liquid meal. When correlating subjective hunger ratings
with brain activation, they did not find significant group results. It is possible that
the incentive salience of food stimuli is such a strong driver of brain activation in
obese participants that hunger fails to further modulate activation in this group.
This may also account for why hunger is a poor predictor of prospective food
consumption in this group. For normal-weight participants, it appears that the
interoceptive cues of physiological hunger may be more important for modulating
response to foods, especially low-calorie foods and food cues.

“Liking” and “Wanting”

“Liking” in the context of food refers to the hedonic value or pleasure
derived from food, whereas “wanting” is related to incentive salience or the
motivational value of a food which drives food consumption (e.g., Berridge et al.,
2008). These two psychological constructs, in conjunction with learning, are
thought to function together in reward-related contexts. “Liking” and “wanting” are thought to be mediated by overlapping, but also separable neural substrates (e.g., Berridge et al., 2008). It is important to note that Berridge (2007) emphasizes that the subjective experiences of “liking” and “wanting” (which we measured in this study) are the interpretation, probably mediated by cortical processes, of more basic “liking” and “wanting”, thought to be mediated by largely subcortical processes. Studies have found that “liking”, largely an opioid-mediated process, is coded in OFC, ACC, insula, AMYG, NAc/ventral striatum, ventral pallidum, and VTA (see Berridge et al., 2008 for a comprehensive review).

We measured “liking” as the subjective assessment of the pleasantness of both high- and low-calorie food images. In response to high-calorie food images, normal-weight controls had widespread reward-related activation predicted by “liking” ratings in 10 of 13 ROIs with bilateral activation in 8 ROIs. Left insula was the only region in this same group in which activation was predicted by “liking” ratings in response to low-calorie foods. The obese group had activation in only two small foci, right Lat OFC (7 voxels) and right ventral pallidum (8 voxels), predicted by increased “liking” ratings in the high-calorie food condition and no regions activated in the low-calorie food condition. There were no group differences found for the low-calorie food condition. Group comparisons for the high-calorie food condition, on the other hand, resulted in a positive relationship between “liking” ratings and activation in normal-weight controls compared to obese participants in left Lat OFC, bilateral Med PFC, left ACC, bilateral insula,
right caudate, and left putamen. The obese group had a greater regression coefficient in only one region, right ventral pallidum, compared to normal-weight controls. The ventral pallidum appears to encode the hedonic value or “liking” of a stimulus in conjunction with the NAc (Berridge and Robinson, 2003; Berthoud, 2004; Smith and Berridge, 2005, 2007). Overall, these results indicate that subjective “liking” appears to modulate reward-related activation in response to high-calorie foods in more regions in normal-weight compared to obese individuals.

“Wanting”, on the other hand, appears to be largely a dopamine-mediated mesocorticolimbic process. Studies have shown “wanting” is coded in VTA, NAc, AMYG, OFC, insula, HIPPO, and ventral pallidum (see Berridge, 2007 for a comprehensive review). In this study, “wanting” was measured as the subjective report of desire (appetite) for the various high- and low-calorie food stimuli presented to participants. Within-group analyses resulted in similar findings for the obese group for high-calorie foods and the normal-weight controls for low-calorie foods. Obese participants had a positive relationship between “wanting” ratings and activation in response to high-calorie foods in left Lat OFC, bilateral Med PFC, bilateral ACC, and left caudate. There was a larger regression coefficient for the low-calorie foods contrast in only one region (right ACC), which overlapped with the high-calorie contrast findings, in the obese group. In response to low-calorie foods, controls had increased activation in right Lat OFC, bilateral Med PFC, bilateral insula, and right putamen predicted by greater “wanting” ratings. High-calorie foods resulted in a larger regression coefficient for
only one region (right insula), which was also activated to low-calorie foods, in the controls. Between-group analyses for the high-calorie food contrast resulted in greater activation in bilateral Med PFC and left caudate in obese compared to normal-weight participants. There were no regions with larger regression coefficients in controls compared to the obese group. The only group difference in the low-calorie food condition was a larger regression coefficient for right ACC in the obese group compared to the controls.

Our results suggest that “wanting” or incentive salience, as opposed to “liking” or hedonic value, is a better predictor of the greater reward system activation in response to foods, especially high-calorie foods, in obese compared to healthy-weight individuals. This is consistent with the psychological literature indicating that the reinforcement value (i.e., “wanting”), but not subjective pleasantness (i.e., “liking”) of foods, especially high-calorie foods, is heightened in obese individuals, and that this may lead to increased consumption of these foods and subsequent weight gain in this population (Epstein and Leddy, 2006; Epstein et al., 1996; Johnson & Wildman, 1983). It is important to note that we found there to be a large degree of overlap in regions showing activation associated with “liking” and “wanting.” This may indicate that the subjective experiences of “liking” and “wanting” may not reflect the underlying differences in neural representation of the more basic “liking” and “wanting” components of reward. We will briefly discuss the possible shared “liking” and “wanting” roles of a few of these ROIs, but more extensive discussion can be found in Stoeckel et al. (2008). The NAc has been conceived as an interface between areas
mediating reward and the energy balance mechanisms of the hypothalamus (e.g., Kelley, 2004). In a circuit involving reciprocal connections with the ventral pallidum, the NAc is involved in the mediation of emotional and motivational responses to food (Smith & Berridge, 2007). The ventral pallidum receives projections from NAc and also VTA and may function as the interface structure at the end of the mesocorticolimbic reward path before signals are sent to the thalamocortical entry loop or motor outputs in brainstem (Berridge, 2007). There is evidence that Med PFC and rostral ACC process the relative value of an anticipated reward and use that information in mediating action selection between competing responses (Amodio & Frith, 2006; Marsh et al., 2006).

*Interoceptive and Exteroceptive Cues*

Hunger is an individual’s interpretation of internal physiological cues reflecting a need for food and has been shown to modulate response to food stimuli (e.g., Fantino et al., 1984; Stoeckel et al., 2007). Marcelino et al. (2001) have demonstrated that appetite is determined by subjective hunger, and that the higher the appetite rating, the more likely people are to initiate eating. However, hunger is not the only factor triggering food intake and in some cases, external factors have been sufficient to stimulate food consumption despite an internal state of satiation (e.g., Cornell et al., 1989; Tuomisto et al., 1998). Exteroceptive factors modulating food intake include perceived time of day, the sight or smell of food, anticipation of the pleasurable consequence of eating, and craving for a particular food or type of food (e.g., Schachter, 1968; Tuomisto et al., 1998).
Original research by Schachter in the 1960’s (e.g., Schachter, 1968; Schachter, 1971; Schachter and Rodin, 1974) found that compared to normal-weight controls, food intake behavior in obese individuals was more heavily influenced by exteroceptive factors such as the salience of foods. In obese individuals, as much as 40% of meal initiation has been attributed to exteroceptive factors such as the time of day whereas only 21% was attributed to subjective hunger (Tuomisto et al., 1998). It has been proposed that obese individuals may not recognize physiological cues of hunger as well as those of normal-weight, possibly due to inadequate learning of physiological cues for hunger (Canetti et al., 2002).

In our study, we found that hunger (an interoceptive factor) appeared to be more important for modulating the response to food cues, especially low-calorie food cues, in normal-weight participants compared to our obese participants. On the other hand, the greater reward-related activation to high-calorie foods we observed in obese participants compared to controls seemed to be explained by the incentive salience of these stimuli, which may lead to hyperphagia, especially related to these high-calorie foods, and subsequent obesity. In normal-weight controls, reward-system activation in response to high-calorie foods appeared to be more closely related to the hedonic value of these stimuli, which has not been shown to be a good predictor of prospective food consumption. Responses to both interoceptive and exteroceptive cues differentiated obese and normal-weight participants in our study, but the direction of these group differences changed depending on whether we were measuring hunger or subjective reward.
value. Hunger (an interoceptive factor) appeared to modulate reward-related brain activation in response to foods, especially low-calorie foods, more in normal-weight controls than obese participants. Subjective reward value (specifically incentive salience), on the other hand, appeared to modulate reward-related brain activation in response to foods, especially high-calorie foods, more in obese compared to normal-weight participants. We hypothesize that interoceptive cues may be less effective at driving eating behavior in obese individuals and that these individuals might rely more on the learned rewarding properties of food cues, especially high-calorie food cues, in the environment to regulate food intake behavior.

Caveats and summary

Many of the important caveats of this study are similar to those mentioned in Stoeckel et al. (2008). Most importantly, we studied a rather homogeneous group of obese participants – young-adult women without eating disorders all imaged in the follicular phase of the menstrual cycle. Further research is necessary to determine the generality of our results. Additionally, we had relatively smaller sample sizes for the valence and appetite ratings analyses, which likely limited our power to detect other significant differences between obese and normal-weight participants.

In summary, our neuroimaging study found differences in activation modulated by both interoceptive (hunger) and exteroceptive (incentive salience)
factors in obese individuals compared to controls in regions of the brain thought to mediate motivational, affective, and associative processing of foods and food cues. There was relatively less hunger-modulated activation to foods, especially low-calorie foods, in obese individuals compared to controls. However, the exteroceptive results were not as straightforward because we found a different pattern of results for the appetite and valence data. There was relatively more activation modulated by foods’ incentive salience, especially for high-calorie foods, in obese compared to normal-weight individuals. On the other hand, activation was more related to valence ratings, for high-calorie foods only, in the controls than in the obese participants. However, as mentioned previously, it appears that incentive salience relates more closely to food intake than valence does. The implications of these findings might be that interoceptive cues, like hunger, may be less effective at regulating eating behavior in obese individuals, and that heightened incentive salience of food cues, especially high-calorie food cues, in the environment may overwhelm these relatively weaker homeostatic mechanisms, leading to hyperphagia and increased weight gain in these individuals. These findings might also provide additional targets for anti-obesity treatments such as psychological or pharmacotherapy that might dampen the mechanisms underlying heightened incentive salience of high-calorie foods, or mindfulness-based therapies that might increase awareness of interoceptive cues, such as hunger.
References


EFFECTIVE CONNECTIVITY OF A REWARD NETWORK IN OBESE INDIVIDUALS

by

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Abstract

Exaggerated reactivity to food cues, especially high-calorie food cues, in obese women appears to be mediated in part by a hyperactive reward system that includes the nucleus accumbens (NAc), amygdala (AMYG), and orbitofrontal cortex (OFC). The present study used fMRI to investigate whether differences in reward-related brain activation in response to high- and low-calorie food images between 12 obese and 12 normal-weight women can be explained by changes in the functional interactions between key reward network regions. A two-step path analysis/General Linear Model (GLM) approach was used to test whether there were group differences (obese vs. normal-weight participants) in network connections between NAc, AMYG, and OFC in response to high- and low-calorie food images. There was abnormal connectivity in the obese group in response to both high- and low-calorie food cues compared to normal-weight controls. Compared to controls, the obese group had a relative deficiency in AMYG projections modulating activation in both OFC and NAc, but excessive influence of the OFC projections modulating activation in NAc. The deficient projections from AMYG might relate to suboptimal modulation of the affective/emotional aspects of a foods’ or food cues’ reward value, whereas increased OFC → NAc connectivity might indicate a heightened drive to consume foods leading to hyperphagia and increased weight gain. Thus, it is possible that not only greater activation of the reward system, but also differences in the interaction of regions in this network may contribute to the relatively increased motivational value of foods in obese individuals.
Keywords: connectivity, food cues, obesity, reward system
The etiology of obesity appears to be explained, in part, by exaggerated reactivity to high-fat, energy dense foods (e.g., Berthoud and Morrison, 2008). The mechanism for the heightened motivational value of these foods in obese individuals may be a hyperactive reward system, which includes the nucleus accumbens/ventral striatum (NAc), amygdala (AMYG), and orbitofrontal cortex (OFC). A previous functional magnetic resonance imaging (fMRI) study found increased activation of these regions in response to high-calorie food images in obese compared to normal-weight individuals (Stoeckel et al., 2008; Fig. 1). Stimuli associated with high-calorie foods may trigger excessive motivation for non-homeostatic eating of these types of foods (Berthoud, 2004a, 2004b; Mela, 2006). This excessive non-homeostatic desire to consume foods has been termed incentive salience or “wanting” and appears to be largely regulated via the mesocorticolimbic dopamine system, which includes NAc, AMYG, and OFC (e.g., Berridge, 2007).

Most human fMRI studies use a mass univariate statistical analysis approach to discern the functional characteristics of different macroscopic brain regions. Investigators often integrate information about the functional specialization of a group of regions to explain how these regions might interact to perform a given function. However, the only empirically based conclusions that are valid relate to the magnitude and extent of activation in a given set of brain regions, not how these regions functionally interact. Connectivity analyses allow researchers to investigate how networks of brain regions interact to perform cognitive and behavioral functions (e.g., Horwitz, 2003). It is important to note
that inferences from traditional activation studies do not directly transfer to connectivity studies. That is, there may be measured differences in the magnitude of brain activation between groups, but no group differences in connectivity, and vice versa (e.g., Mechelli et al., 2007).

Path analysis, a subset of structural equation modeling, is a multivariate, hypothesis-based approach applied to functional neuroimaging to investigate the directional relationship between a given set of brain regions and their connections (McIntosh et al., 1994a, 1994b). Compared to data-driven connectivity approaches, path models are developed based on a priori hypotheses and assume a causal structure, where A \( \rightarrow \) B means changes in region A are hypothesized to cause changes in region B (e.g., Penny et al., 2004). Brain regions in a network model are typically selected based on previous functional neuroimaging studies, and connections between these regions are usually defined based on the known neuroanatomical connections, mostly from animal literature, assuming homology in brain regions between species (e.g., Penny et al., 2004). The parameter estimate values calculated using path analysis represent the quantification of the directional pathways between regions in the model. Parameter estimate values or path coefficients can then be used to make comparisons between connections within subjects in response to changes in task conditions, or between subjects and groups within the General Linear Model (GLM) framework (e.g., Kim et al., 2007; Protzner and McIntosh, 2006).

The NAc, AMYG, and OFC function together as part of the reward system. There has been a call to investigate this network and its pathways, in particular,
and how they are functioning to code for the rewarding effects of foods in a manner that would promote obesity (Berthoud, 2004b; 2006). There are strong anatomical connections between NAc, AMYG, and OFC (see Fig. 2), from AMYG → OFC (Cavada et al., 2000; Heimer and Van Hoesen, 2006; Kalivas and Nakamura, 1999; Kringelbach and Berridge, 2008; Petrides, 2007; Rempel-Clower, 2007; Schmidt et al., 2005), AMYG → NAc (Heimer and Van Hoesen, 2006; Kalivas and Nakamura, 1999; Schmidt et al., 2005), and OFC → NAc (Cavada et al., 2000; Cohen et al., 2005; Heimer and Van Hoesen, 2006; Kalivas and Nakamura, 1999; Kringelbach and Berridge, 2008; Morecraft et al., 1992; Petrides, 2007; Rempel-Clower, 2007; Schmidt et al., 2005). Although it is clear that NAc, AMYG, and OFC are more strongly activated in obese compared to normal-weight controls when viewing food images, particularly high-calorie food images (Stoeckel et al., 2008), it is uncertain whether activation in these regions relates to some common underlying reward process (e.g., incentive salience) or whether there are different processes (e.g., hedonics or learning) that account for this activation pattern. The NAc, AMYG, and OFC each have numerous functional properties. Together, AMYG and OFC may mediate the associative processes whereby food-related stimuli acquire incentive salience or other motivational properties (e.g., Berridge, 2007; Holland and Petrovich, 2005), but both also code for hedonic value, AMYG via bottom-up and OFC via top-down processes (Kringelbach and Berridge, 2008). The OFC may encode multimodal sensory representations of food and food cues (Berthoud, 2004a). The NAc/ventral striatum functions as an interface between reward-related
processing, homeostatic mechanisms, and motor output (e.g., Kelley, 2004), but may also code for reward value (O'Doherty et al., 2006).

In this study, we used fMRI and a two-stage path analysis plus GLM approach to investigate the interactions of key reward structures (NAc, AMYG, and OFC) in a simple network to determine whether these structures function together in response to images of high- and low-calorie foods differently in obese and normal-weight individuals. We expected to find effective connections between brain regions as specified in our model in both obese participants and normal-weight controls in response to high- and low-calorie food images. In addition, we expected to find a number of altered effective connections in our obese group that might help explain why foods, especially high-calorie foods, have increased motivational potency for these individuals.
Fig. 1. Greater activation found in obese than in control participants to high-calorie foods > cars in (A) left Lat OFC (axial view). Greater activation found in obese than in control participants to high-calorie > low-calorie foods in (B) left AMYG (coronal view) and (C) right NAc (coronal view), enhanced view of image on right. Activation is overlaid on the SPM2 single-subject T1 template.

**Materials and methods**

With the exception of the section discussing the methods of path analysis, the information below is provided in greater detail in Stoeckel et al. (2008).

*Participants*

Participants were 12 obese (Body Mass Index, BMI, = 30.8 – 41.2) and 12 normal-weight (BMI = 19.7 – 24.5) right-handed women recruited from the University of Alabama at Birmingham community. There were no group
differences on mean age (obese: 27.8, SD = 6.2; control: 28.0, SD = 4.4),
ethnicity (obese: 7 African-American, 5 Caucasian; control: 6 African-American, 6
Caucasian), education (obese: 16.7 years, SD = 2.2; control: 17.2, SD = 2.8), or
mean day of the menstrual cycle (obese: day 6.8, SD = 3.1, control: day 5.7, SD
= 3.3, all in the follicular phase). Individuals were excluded based on criteria
including a positive eating disorder history, active dieting or participating in a
weight-loss program, or weight > 305 pounds (138 kg) with girth > 64 inches (163
cm), the latter due to scanner limitations.

Stimuli

The stimuli used during the imaging session consisted of 252 color
pictures from a previous study (Stoeckel et al., 2008), all of consistent size,
resolution, and luminance. The 168 food images were subdivided into low-calorie
and high-calorie categories, each consisting of 84 unique images. Low-calorie
food images consisted of such low-fat items as steamed vegetables and broiled
fish. High-calorie foods were primarily items high in fat such as cheesecake or
pizza. Control stimuli consisted of car images of a wide range of makes, models,
ages, and colors.

Procedure

After thorough screening to validate BMI, eating disorder diagnosis, and
other study criteria, participants were scheduled for the fMRI session. They were
instructed to eat a normal breakfast between 7-8 A.M. but to skip lunch and
consume only water so that they had fasted for approximately 8-9 h before being imaged between 3-5 P.M. There were no group differences on subjective hunger ratings.

While participants were in the magnet, visual stimuli were presented in a block design format, with a total of six 3:09 min runs per imaging session. Each run consisted of two 21 s epochs each of cars (C), low-calorie foods (LC), and high-calorie foods (HC) pseudorandomly presented to the participants. Within each 21 s epoch of food or car images, seven individual images were presented for 2.5 s each followed by a 0.5 s gap each, separated by 9 s of a gray blank screen with a fixation cross. Each run consisted of 63 volumes for a total of 378 volumes across six runs, of which 84 volumes were acquired during each of the car, low-calorie food, and high-calorie food exposures. The visual images were presented by a laptop using VPM software (Cook et al., 1987). Images were projected onto a screen behind the participant’s head and viewed via a 45° single-surface rear-projecting mirror attached to the head coil. Participants were financially compensated for their participation. All procedures were reviewed and approved by UAB’s Institutional Review Board for Human Use.

**MRI acquisition and processing**

Functional MRI data were acquired using a Philips Intera 3T ultra-short bore magnet equipped with a SENSE head coil. Images were collected using a single-shot T2*-weighted gradient-echo EPI pulse sequence. We used TE = 30 msec, TR = 3 sec, and an 85° flip angle for 30 axial slices 4 mm thick with a 1
mm interslice gap, a scan resolution of 80 x 79, reconstructed to 128 x 128, and with a 230 x 149 x 230 mm FOV. The first four scans were discarded to allow the magnet to achieve steady-state magnetization.

Data were preprocessed (motion correction, normalization to the MNI coordinate system using the SPM2 EPI template, and smoothed with a 6 mm FWHM Gaussian filter) using the SPM2 software package (Wellcome Dept. Imaging Neuroscience, London, UK). No data sets failed to meet the movement inclusionary criteria, which were that movement before correction was < 2 mm in translational movement and < 2° in rotational movement (details in Stoeckel et al., 2008).

Data analysis

fMRI data. Block-design blood oxygen level dependent (BOLD) responses were analyzed within the context of the General Linear Model on a voxel by voxel basis as implemented in SPM2 (Friston et al., 1995). The time course of brain activation was modeled with a boxcar function convolved with the canonical hemodynamic response function (HRF) and a temporal derivative function. The data were high-pass filtered (1/128 Hz) to remove low frequency drifts. A first order autoregressive model was also implemented to correct for autocorrelations in the error term of the fMRI model.

A two-stage random-effects procedure was used for the statistical analysis to account for both within-subject and between-subject variability. First, the fMRI data from each individual participant were used to generate statistical contrasts
of the parameter estimates in order to test the differences between the time points corresponding to the high-calorie and low-calorie foods. Results of a previous study (Stoeckel et al., 2008) found group differences in patterns of reward-related activation, with the obese group exhibiting greater activation to high-calorie foods and the control group to low-calorie foods. The food > control stimuli contrast was then entered into second-level one-sample t-test analyses for the within-group comparisons to localize the group maxima for our regions of interest (ROI): bilateral NAc, AMYG, and OFC (p < .05, uncorrected; Mechelli et al., 2007).

ROI’s were defined using the WFU Pickatlas and the AAL and Talairach Daemon atlases (Lancaster et al., 2000; Maldjian et al., 2004; Tzourio-Mazoyer et al., 2002). NAc was unavailable in these libraries, so we drew a three-dimensional sphere with the WFU Pickatlas centered at a voxel location determined by averaging voxel location dimensions from relevant fMRI studies (Aron et al., 2005; Menon and Levitin, 2005; O’Doherty et al., 2002). The classification of activated voxels was verified by using the WFU Pickatlas and visual inspection of the data using a human brain atlas (Mai et al., 2004).

Path analysis

Path analysis was used to determine the strength and direction of the relationship (connections) between observed variables (ROIs) estimated using simultaneous regression equations via maximum likelihood estimation. Path analysis, often referred to as structural equation modeling, is the most common
modeling approach used in functional neuroimaging (Schlosser et al., 2005). We used a two-step path analysis/GLM approach, following a similar method as Kim et al. (2007). For each participant: (1) ROIs were selected to include in the model, (2) the time series data were partitioned according to task conditions, accounting for the hemodynamic lag, (3) summary data were extracted for each condition for each ROI, (4) a model was designated that specified the interactions of the ROIs, (5) the variance-covariance (time series data X ROI) matrix for each condition was calculated, and (6) the path coefficients for the connections between ROIs in the models were estimated via maximum likelihood estimation. Repeated-measures ANOVA was then used to determine within-group (i.e., condition) and between-group differences in the model connections using the path coefficients from the models for each individual.

*Model specification.* The regions included in the model (OFC, AMYG, and NAc) are part of what has been termed the “motive circuit” (Pierce and Kalivas, 1997) involved in reward-related functioning likely via the mesocorticolimbic dopamine system (Berridge, 2007; Jentsch and Taylor, 1999; Kalivas and Volkow, 2005; Kolb, 1999; Pierce and Kalivas, 1997; Robinson and Berridge, 2003; Simansky, 2005; Volkow et al., 2003; Zahm, 2000). The connections in the model were defined based on the known anatomical connectivity of the structures in this network (Heimer and Van Hoesen, 2006; Kalivas and Nakamura, 1999; Berridge and Kringelbach, 2008; Petrides, 2007; Rempel-Clower, 2007; Schmidt et al., 2005; Fig. 2). In order to estimate reliable path
coefficient values, the model was constrained to be recursive (i.e., no reciprocal paths were included in the model).

The same path model was constructed for each subject. To allow for inter-subject variability, we defined the exact coordinates of each ROI for each hemisphere from the local maxima of each participant’s statistical map within 12 mm of the group maxima (within the same anatomical region) resulting from the foods > cars contrast (p < .05, uncorrected; Mechelli et al., 2007). The MNI coordinates of the regions were NAc, left (x, y, z): -6, 10, -10 [controls] and -10, 14, -6 [obese]; NAc, right, (x, y, z): 6, 10, -10 [controls] and 6, 12, -10 [obese]; AMYG, left (x, y, z): -26, -2, -20 [controls] and -20, 0, -24 [obese]; AMYG, right (x, y, z): 22, 0, -20 [controls] and 24, 2, -24 [obese]; OFC, left (x, y, z): -22, 36, -10 [controls] and -22, 30, -14 [obese]; OFC, right (x, y, z): 26, 36, -14 [controls] and 26, 30, -4 [obese]. For each region, the principal eigenvariate of the time series was extracted from a 4-mm sphere centered at the subject-specific local maximum. The principal (i.e., 1st) eigenvariate is a summary measure, similar to a weighted mean robust to outliers, based on the variance of all the voxels included with the 4-mm sphere.

The regional time series data (principal eigenvariate values) were then separated into two data sets: time points associated with (1) the high-calorie foods and (2) the low-calorie foods. To account for the hemodynamic lag, we assumed a 6 s (2 TR) physiological delay between the onset and offset of our two conditions and adjusted the data we extracted accordingly (Honey et al.,
This resulted in two 84 (time points) X 6 (ROIs) matrices of data for each condition (high- and low-calorie foods) for each participant.

*Path parameter estimates.* A path model was fit to the data matrix for both the high-calorie and low-calorie foods independently for each participant. The free path coefficients were estimated by minimizing the discrepancy between a correlation matrix observed from the fMRI data and a correlation matrix predicted by the model using LISREL software. The standardized parameter estimates (similar to $\beta$s in regression) or path coefficients for each connection (AMYG $\rightarrow$ OFC, OFC $\rightarrow$ NAc, and AMYG $\rightarrow$ NAc) within each hemisphere (left and right) from both models (high- and low-calorie foods) for each participant were imported into SPSS for subsequent analyses. A repeated-measures ANOVA was used to determine group differences between obese and control participants for high- and low-calorie foods (category) in the left and right hemisphere (laterality) for each path. As this was an exploratory study, we also tested for the significance of path coefficients despite non-significant omnibus models. For each group, one sample t-tests were used to test whether the path coefficients for each hemisphere were significantly different than zero, indicating connectivity as specified in the high- and low-calorie food models. Pairwise comparisons were used to test the differences in path coefficients for each hemisphere (left and right) for within-group (high-calorie vs. low-calorie foods) and between-group comparisons (obese vs. controls) for high-calorie and low-calorie foods, independently. Paired t-tests were used for within-group comparisons and independent samples t-tests were used for between-group comparisons.
Results

All the estimated path coefficients were significant for the obese group and controls for both hemispheres in both the high- and low-calorie food models, indicating that the regions in this network interacted as specified in the model ($p < 0.001$; Table 1).
Table 1. The path coefficients for the connections tested in the reward model for the high-calorie food and low-calorie food conditions for the obese and normal-weight groups.

<table>
<thead>
<tr>
<th>Path Connections</th>
<th>HC (Obese)</th>
<th>HC (Controls)</th>
<th>LC (Obese)</th>
<th>LC (Controls)</th>
</tr>
</thead>
<tbody>
<tr>
<td>L AMYG → L OFC</td>
<td>0.64 ± 0.10</td>
<td>0.86 ± 0.03</td>
<td>0.65 ± 0.09</td>
<td>0.80 ± 0.03</td>
</tr>
<tr>
<td>R AMYG → R OFC</td>
<td>0.65 ± 0.09</td>
<td>0.86 ± 0.04</td>
<td>0.62 ± 0.07</td>
<td>0.82 ± 0.03</td>
</tr>
<tr>
<td>L AMYG → L NAc</td>
<td>0.35 ± 0.07</td>
<td>0.60 ± 0.04</td>
<td>0.37 ± 0.05</td>
<td>0.52 ± 0.05</td>
</tr>
<tr>
<td>R AMYG → R NAc</td>
<td>0.36 ± 0.06</td>
<td>0.45 ± 0.06</td>
<td>0.32 ± 0.04</td>
<td>0.39 ± 0.06</td>
</tr>
<tr>
<td>L OFC → L NAc</td>
<td>0.54 ± 0.06</td>
<td>0.31 ± 0.05</td>
<td>0.52 ± 0.05</td>
<td>0.35 ± 0.05</td>
</tr>
<tr>
<td>R OFC → R NAc</td>
<td>0.52 ± 0.08</td>
<td>0.49 ± 0.06</td>
<td>0.57 ± 0.05</td>
<td>0.52 ± 0.06</td>
</tr>
</tbody>
</table>

L = left, R = right, HC = high-calorie foods, LC = low-calorie foods. All other conventions as in the paper.

**Omnibus analyses**

**AMYG → OFC**

There was a main effect of group such that the connectivity from AMYG → OFC was greater for controls (0.84 ± 0.07) compared to the obese group (0.64 ± 0.07), indicating a relatively stronger directional relationship in brain activation between these structures in response to foods in controls (F [1,22] = 4.46, p = 0.046). There were not significant group by category or group by laterality interactions, although the group by category by laterality interaction was significant at the trend level (p = 0.066).

**AMYG → NAc**

There was a main effect of group for the AMYG → NAc connection such that there was greater connectivity for the controls (0.49 ± 0.05) compared to the
obese group (0.35 ± 0.05). There were not significant group by category or group by category by laterality interactions, although the group by laterality interaction was significant at the trend level (p = 0.09).

OFC → NAc

There was no main effect of group for the OFC → NAc connection, although there was a group difference at the trend level (F [1,22] = 3.70, p = 0.067) indicating greater connectivity for the obese group (0.53 ± 0.06) compared to the controls (0.41 ± 0.06). There were not significant group by category or group by category by laterality interactions, although the group by laterality interaction was significant at the trend level (p = 0.059).

Pairwise comparisons and simple interactions

Between-group comparisons: obese vs. controls for high- and low-calorie foods

Controls > Obese (High-calorie foods). The path coefficient from left AMYG → left NAc was greater in the controls compared to the obese group within the high-calorie food condition (p = 0.003; Fig. 3a). There were significant differences from AMYG → OFC bilaterally (left: p = 0.05, right: p = 0.05; Fig. 3a).

Controls > Obese (Low-calorie foods). The path coefficients from right AMYG → right OFC (p = 0.02) and left AMYG → left NAc (p = 0.046) were greater in the controls compared to obese group within the low-calorie food condition (Fig. 3b).
Obese > Controls (High-calorie foods). The path coefficient from left OFC → left NAc was greater in the obese group compared to controls within the high-calorie food condition, p = 0.007 (Fig. 3a).

Obese > Controls (Low-calorie foods). There was a similar group difference from left OFC → left NAc within the low-calorie food condition (p = 0.03) as in the high-calorie food comparison (Fig. 3b).

Within-group comparisons: high- vs. low-calorie food conditions

Controls. The path coefficients from AMYG → OFC bilaterally (left: p = 0.007, right: p = 0.002) were significantly greater for the high- vs. low-calorie food category comparison in the controls. There were no path coefficients greater during the low- compared to high-calorie food condition within this group (Fig. 4).

Obese. There were no significantly different path coefficients between the high- and low-calorie food conditions within the obese group.
Fig. 3. Group comparisons (obese vs. controls) related to the path coefficients for the (A) high-calorie foods and (B) low-calorie foods. Thicker arrows indicate significant or trend-level differences. OB = obese, CTRL = controls. All other conventions as mentioned previously.
Fig. 4. Food category (high-calorie foods vs. low-calorie foods) comparisons within the control group. Thicker arrows indicate significant or trend-level differences. HC = high-calorie foods, LC = low-calorie foods. All other conventions as mentioned previously.

Between-group comparisons: high- vs. low-calorie food conditions

None of the group x food category interactions were significant for the comparisons in either hemisphere. There were trends for greater differences between high- and low-calorie foods from left AMYG → left OFC (p = 0.068) and left AMYG → left NAc (p = 0.072) in controls compared to the obese group.
Discussion

Previous research has shown that food cues, especially those associated with high-calorie foods, trigger hyperactivity in brain regions including NAc, AMYG, and OFC thought to mediate or at least code for motivational and emotional processes in obese individuals (Stoeckel et al., 2008). In the present study, we tested whether there were differences in network connections between NAc, AMYG, and OFC in response to high- and low-calorie food images within and between obese and normal-weight groups. We found aberrant connectivity in the obese group in response to both high- and low-calorie food cues compared to normal-weight controls. Specifically, it appears that the obese group had a relative deficiency in AMYG modulation of activation in both OFC and NAc, but excessive influence of the OFC projections modulating activation in NAc. Thus, it is possible that not only greater activation of the reward system, but also differences in the interaction of regions in this network may contribute to the relatively increased motivational value of foods in obese individuals.

The reward model

All path connections were significant for both high- and low-calorie food models in both the obese group and normal-weight controls, indicating NAc, AMYG, and OFC were functioning together as specified in the model. This is consistent with the known anatomical connections between these regions (Cavada et al., 2000; Cohen et al., 2005; Heimer and Van Hoesen, 2006; Kalivas
and Nakamura, 1999; Kringelbach and Berridge, 2008; Morecraft, 1992; Petrides, 2007; Rempel-Clower, 2007; Schmidt et al., 2005). This network consisting of the NAc, AMYG, and OFC is innervated by the ventral tegmental area, which releases dopamine to this circuit in response to motivationally salient events (Berridge and Robinson, 2003; Kalivas and Volkow, 2005; Schmidt et al., 2005). However, the projections between NAc, AMYG, and OFC as illustrated in Fig. 2 are glutamatergic (Kalivas and Volkow, 2005; Schmidt et al., 2005).

This NAc, AMYG, and OFC reward network is a subcircuit of a larger “motive circuit” thought to function in order to activate and direct behavior in response to motivationally-relevant stimuli (Kalivas and Volkow, 2005; Pierce and Kalivas, 1997). The NAc, AMYG, and OFC, in particular, have important reward-related functions that likely contribute to both general and food-specific motivational processes (Berridge, 2007; Berthoud, 2004a; Jentsch and Taylor, 1999; Kalivas and Volkow, 2005; Kolb, 1999; Pierce and Kalivas, 1997; Robinson and Berridge, 2003; Simansky, 2005; Volkow et al., 2003; Zahm, 2000). The AMYG appears to be involved in motivationally-relevant associative processes (Petrovich and Gallagher, 2007; Petrovich et al., 2005). In addition to coding for more general affective and motivational properties, the AMYG may mediate the specific properties of food-related stimuli (Balleine and Killcross, 2006). The OFC appears be a key region for translating reward value into hedonic experience (Kringelbach, 2005), processing the temporal and certainty characteristics of reward (Cardinal, 2006), and is also involved in motivation-related learning processes in conjunction with AMYG (Everitt et al., 1999; Parkinson et al., 2000).
The OFC shows multimodal responses to food cues (Rolls, 2005) and has been referred to as the ‘secondary taste area’, following gustatory processing in insular cortex (Berthoud, 2004a). The NAc/ventral striatum may have numerous reward-related roles. It has been conceptualized as the ‘limbic-motor’ interface (Mogenson, 1980) and appears to be involved in processing related to Pavlovian conditioning, incentive salience, and reward availability, value, and context (Bradberry, 2007; Cardinal et al., 2002; Day et al., 2007). This region, in conjunction with ventral pallidum, via opioid-mediated mechanisms, may also code for hedonic value (Berridge and Robinson, 2003; Berthoud, 2004a; Smith and Berridge, 2005, 2007). For food reward, the NAc/ventral striatum may integrate homeostatic and non-homeostatic signals to modulate motivational state (Kelley et al., 2005). This region might also code for the relative reward value of available food stimuli (O’Doherty et al., 2006).

It is important to note that this is the first human connectivity study using functional neuroimaging to measure the interaction of brain regions in a reward network. Most of what is written about the functional relationships between the regions in our model comes from animal research and assumes homology in these areas between species. By design, this study is exploratory in nature and our interpretations of the results and their relationship to obesity are speculative, with the intention of provoking more questions as opposed to resolving unanswered questions. Before discussing the implications of our findings, we will review some limitations in this study, mostly related to methodological constraints.
(1) Specifying a model using path analysis in fMRI can be a challenge because the number and combination of connections between regions increase substantially with every additional region included in the model, which makes estimating these path coefficients reliably and interpreting the findings more difficult. For instance, in this study with 3 regions per hemisphere (6 regions total), there are \( k = \frac{N(N + 1)}{2} = 21 \) degrees of freedom per data set (\( k = 42 \) degrees of freedom for the two models tested) allotted to estimate the effects of interest. Twelve degrees of freedom are used to estimate the variances associated with each region in both models (6 regions per model x 2 models). With a minimum of 5 data points necessary to estimate the parameter values for each path in the model reliably (Bentler and Chou, 1987), this leaves a maximum of 30 estimable paths for two models with 6 regions each (15 estimable paths per model). This limits the complexity of the model that can be tested using path analysis and is one reason we chose not to include interhemispheric connections in our model.

(2) We chose the two-stage path analysis / GLM approach in order to directly test for group differences between connections in a hypothesized model and were not as interested in comparing the fit of the model between groups per se. This approach is different from the traditional fMRI and path analysis methodology termed the “stacked model approach”, comparing model fit between tasks or groups (McIntosh, 1994). However, Protzner and McIntosh (2006) recently reported that reliable parameter estimates could be generated using path analysis even when a model is not a sufficient fit to the data.
(3) Another limitation of this study relates to the power to detect differences between the path coefficients estimated in our models due to the small sample sizes used for each group. With larger group sizes, our trend level findings would likely have reached statistical significance.

(4) We did not include the ventral tegmental area (VTA), the source of dopamine within the mesocorticolimbic circuit proposed to mediate many of the processes associated with reward (Fields et al., 2007; Hyman, 2007; Schultz, 2006), in our model due to methodological limitations related to BOLD fMRI that make detecting activation in brainstem regions like the VTA difficult (D’Ardenne et al., 2008).

**AMYG → OFC**

When directly comparing the obese and normal-weight groups, we found that the connection from AMYG → OFC was greater in controls than obese participants when the high- and low-calorie food categories were combined. Pairwise comparisons resulted in bilateral differences in a similar direction for high-calorie foods and significant differences in the right hemisphere for low-calorie foods. Within the control group, there was greater AMYG → OFC connectivity bilaterally in the high-calorie compared to low-calorie food condition. The obese group, on the other hand, demonstrated no differences in connectivity when comparing high- and low-calorie foods. There was also a trend for a group x food category interaction in the left hemisphere, indicating relatively greater connectivity for controls especially for high-calorie compared to low-calorie foods.
Basic learning whereby stimuli associated with primary rewards acquire motivational value may occur in the AMYG (Berridge, 2004). The projection from AMYG → OFC may transfer basic motivationally relevant associative information (AMYG) to the OFC, which uses information from the AMYG to determine subjective value and influence subsequent instrumental choice behavior (Cardinal et al., 2002). For ingestive behavior, the AMYG → OFC connection might involve the transfer of information relating to the hedonic value of the food-related stimuli (e.g., Berridge and Kringelbach, 2008). The AMYG may mediate the non-conscious, implicit processing relating to the hedonics associated with the food-related stimuli, whereas OFC may mediate conscious, explicit processing of hedonics and the application of subjective hedonic value to the stimulus (e.g., Berridge and Kringelbach, 2008). A deficient AMYG → OFC connection in the obese participants may indicate suboptimal transfer of basic affective/emotional value regarding foods and food cues important for updating the subjective reward value of these cues to facilitate flexibility in food intake behavior. Without these basic modulatory signals (AMYG), the reward value of foods or food cues (OFC) might not be diminished at the same rate as is normally observed in devaluation following food intake. There might be a diminished decrease in the drive to consume foods following intake in some individuals, which could lead to hyperphagia and increased weight gain. Although there is no direct evidence to support this, Baxter and colleagues (2000) found that macaque monkeys failed to change their behavior during a reward devaluation task after the connection between AMYG and OFC was disrupted.
**AMYG → NAc**

The results for the AMYG → NAc analyses were similar to the AMYG → OFC analyses, indicating that the connection was greater for controls compared to the obese group when the high- and low-calorie food categories were combined. Independent analyses of both high-calorie and low-calorie food models revealed significant left hemisphere differences in a similar direction for both models. There was also a trend for a group x food category interaction in the left hemisphere, indicating relatively greater connectivity for controls especially for high-calorie compared to low-calorie foods. Similar to the AMYG → OFC connection, a deficient connection in the obese from AMYG → NAc might indicate that the basic hedonic signal that serves to modulate the reward value of foods or food cues (AMYG) is not appropriately weighted with other signals (e.g., motivational, homeostatic) before the appropriate ingestive behavior is determined (Zahm, 2006). This might lead to hyperphagia and increased weight gain in a similar way as discussed earlier, only via more unconscious, implicit mechanisms.

**OFC → NAc**

There was a trend for greater connectivity in OFC → NAc in obese compared to controls when the high- and low-calorie food categories were combined. Independent analyses of both high-calorie and low-calorie food models resulted in significant left hemisphere differences in a similar direction for
both models. A greater incoming signal from OFC $\rightarrow$ NAc in obese participants might indicate the transfer of a heightened subjective hedonic value of foods or food stimuli. Addiction investigators have theorized that relatively greater PFC (including OFC) $\rightarrow$ NAc synaptic glutamate transmission may explain increased motivation for drugs and drug-related cues (Kalivas, 2005; Kalivas and Volkow, 2005). There is strong evidence that the rewarding effects of addictive drugs and foods are mediated by a common neural substrate (Kelley and Berridge, 2002; Volkow and Wise, 2005). Thus, in the same way drug addicts have increased drive for drugs and drug cues, some obese individuals may have increased drive for foods and food cues, with heightened drive related to a relatively greater glutamatergic-mediated transmission from OFC $\rightarrow$ NAc.

Conclusions and Summary

In summary, our neuroimaging study found relatively aberrant reward network connectivity in obese individuals compared to controls, with deficient projections from AMYG to OFC and NAc and increased connectivity in OFC $\rightarrow$ NAc in these participants. These results add to the literature showing not only exaggerated reward system activation in response to foods, but also an abnormal interaction between regions in this network that may lead to hyperphagia in obese individuals. In particular, we think overeating in obese individuals might be explained, in part, by two mechanisms: (1) deficient projections from AMYG might indicate suboptimal modulation of the affective/emotional aspects of a foods’ or food cues’ reward value and (2)
increased OFC → NAc connectivity might contribute to a heightened drive to consume foods. Without the appropriate affective/emotional information to signal the devaluation of foods or food cues following food intake, heightened drive may overwhelm homeostatic mechanisms, leading to hyperphagia and increased weight gain. Admittedly, we tested a simple reward network. Further studies are necessary to investigate connectivity in the reward system and how these regions might interact with homeostatic mechanisms in the hypothalamus and brainstem, as well as the cognitive mechanisms of food intake control in the prefrontal cortex. It will also be interesting to determine how individual differences related to reward sensitivity and interoceptive and exteroceptive factors modulate this reward network in order to better understand how reward mechanisms influence ingestive behavior.
References


Kalivas, P.W., 2005. How do we determine which drug-induced neuroplastic changes are important? Nat Neurosci 8, 1440-1441.


SUMMARY

In summary, our neuroimaging study found a greater magnitude of activation to high-calorie foods in obese individuals compared to normal-weight controls within a wide range of brain regions thought to mediate motivational, affective, and reward-related associative mechanisms in response to foods and food cues. We also found abnormal interactions between a subset of these reward-related regions (NAc, AMYG, and OFC) in obese individuals compared to controls. Finally, the obese individuals were further distinguished from controls by a relatively deficiency in hunger-related modulation of reward system activation and greater reward-related modulation related to foods' incentive salience, as opposed to hedonic value.

CONCLUSIONS

These results strongly support a hypothesis based on behavioral studies which proposes that hyperphagia in obese individuals is triggered by the greater potency of stimuli associated with high-calorie foods via exaggerated reactivity of the brain's reward system. In addition, interoceptive cues, like hunger, appear to be less effective at modulating ingestive behavior in obese individuals, and the heightened incentive salience of food cues, especially high-calorie food cues, via non-homeostatic mechanisms may be more important in regulating food intake in obese individuals. Not only exaggerated reward system activation in response to foods, but also an abnormal interaction between a subset of brain regions in this network may also contribute to obesity. Relatively weaker projections from AMYG in obese individuals might indicate suboptimal modulation of the
affective/emotional properties of foods or food cues, and a stronger projection from OFC to NAc might be a good candidate pathway for mediating obese individuals’ heightened drive to consume foods. Without adequate affective/emotional information to modulate the value of foods or food cues, heightened drive may overwhelm the non-homeostatic circuit regulating ingestive behavior and may overwhelm hypothalamic/brainstem-mediated homeostatic mechanisms normally regulating food intake to promote hyperphagia, subsequent weight gain, and obesity.
REFERENCES


APPENDIX

IRB APPROVAL FORM
Form 4: IRB Approval Form
Identification and Certification of Research
Projects Involving Human Subjects

UAB's Institutional Review Boards for Human Use (IRBs) have an approved Federalwide Assurance with the Office for Human Research Protections (OHRP). The UAB IRBs are also in compliance with 21 CFR Parts 50 and 56 and ICH GCP Guidelines. The Assurance became effective on November 24, 2003 and expires on October 26, 2010. The Assurance number is FWA00005960.

Principal Investigator: STOECKEL, LUKE E
Co-Investigator(s):
Protocol Number: X070508010
Protocol Title: Activation of the Reward System in Response to Food Images in Obese and Normal-Weight Individuals: An fMRI Study

The IRB reviewed and approved the above named project on 5-8-08. The review was conducted in accordance with UAB's Assurance of Compliance approved by the Department of Health and Human Services. This Project will be subject to Annual continuing review as provided in that Assurance.

This project received EXPEDITED review.
IRB Approval Date: 5-8-08

Date IRB Approval Issued: 5-8-08

HIPAA Waiver Approved?: N/A

Marilyn Doss, M.A.
Vice Chair of the Institutional Review Board for Human Use (IRB)

Investigators please note:

The IRB approved consent form used in the study must contain the IRB approval date and expiration date.

IRB approval is given for one year unless otherwise noted. For projects subject to annual review research activities may not continue past the one year anniversary of the IRB approval date.

Any modifications in the study methodology, protocol and/or consent form must be submitted for review and approval to the IRB prior to implementation.

Adverse Events and/or unanticipated risks to subjects or others at UAB or other participating institutions must be reported promptly to the IRB.