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ORIGINAL ARTICLE

Neural representations of hunger and satiety in Prader–Willi syndrome

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Objective: To investigate the neural basis of the abnormal eating behaviour in Prader–Willi syndrome (PWS), using brain imaging. We predicted that the satiety response in those with PWS would be delayed and insensitive to food intake.
Design and participants: The design of this study was based on a previous investigation of the neural activation associated with conditions of fasting and food intake in a nonobese, non-PWS group. The findings were used to generate specific hypotheses regarding brain regions of interest for the current study, in which 13 adults with PWS took part (mean $\pm$ s.d. age $= 29 \pm 6$; BMI $= 31.5 \pm 5.1$; IQ $= 71 \pm 8$, six were female).
Measurements: Regional cerebral blood flow was measured using positron emission tomography in three sessions: one following an overnight fast and two following disguised energy controlled meals of similar volume and appearance – one of 1674 kJ (400 kcal) and another of 5021 kJ (1200 kcal). Subjective ratings of hunger, fullness and desire to eat, and blood plasma levels of glucose, insulin, leptin, ghrelin and PYY were measured before and after each imaging session.
Results: The neural representation of hunger, after an overnight fast, was similar to that found in nonobese individuals in the control study. In contrast, after food intake, the patterns of neural activation previously associated with satiety were not found, even after the higher-energy load. Lateral and medial orbitofrontal cortical activation was associated with consumption of the 400- and 1200-kcal meals, respectively. The medial orbitofrontal activation, however, was only found in those who had shown a large percentage change in fullness ratings following the higher-energy meal.
Conclusion: We conclude that there is a dysfunction in the satiety system in those with PWS. These findings suggest that brain regions associated with satiety are insensitive even to high-energy food intake in those with the syndrome. This may be the neural basis of the hyperphagia seen in PWS.
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Keywords: Prader–Willi syndrome; hunger; satiety; hypothalamus; orbitofrontal cortex; positron emission tomography

Introduction

Prader–Willi Syndrome (PWS) results from the absence of expression of the paternal copy of as yet unidentified maternally imprinted gene(s) at the genetic locus 15(q11–13). This syndrome is associated with hyperphagia, leading to morbid obesity if the food environment is unsupervised.[1] The eating behaviour has been characterised as a constant desire to eat, which, together with reduced physical activity in those with PWS,[2] and energy requirements that are 50–75% of the normal,[3] means access to food must be strictly controlled to prevent extreme obesity.

In order to understand the nature and basis of the hyperphagia in PWS, a number of studies have been conducted to quantitatively characterise the behaviour. Zipf and Berntson[4] found that, when presented with a plate of sandwiches for 1 h, the control group all finished eating after 15 min, whereas all but one in the PWS group continued to eat for much longer, in several cases for the whole hour. Using a similar design, Holland *et al*.[5] replicated and extended these findings; the PWS group continued eating for longer and ate on average three times more calories (PWS 1292 vs control 369 kcal) in the hour. This significantly greater amount of food was required in those with PWS to bring about a similar change in feelings of hunger, as

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measured by visual analogue scales, as the control group. Furthermore, it was found that those feelings of hunger in the PWS group were correlated with the extent of change in blood glucose levels, which, in the PWS group only, reached above the accepted physiological range. Using a hidden weighing scale, Lindgren *et al*.[6] found that those with PWS showed nondecelerating eating curves, a slower eating rate and a longer duration of eating compared to both lean and obese children without PWS. Together, these findings suggest that those with PWS have decreased satiation as opposed to extreme hunger, and that the satiety response in those with PWS is at least delayed, if not insensitive, to food.

The mechanisms underlying the eating behaviour in those with PWS are poorly understood. There is some evidence to suggest a hypothalamic basis,[7,8] which was recently supported by a functional neuroimaging study in three individuals with PWS.[9] Shapira *et al*.[9] showed a significant delay in the negative response of the hypothalamus to glucose ingestion in those with PWS, in comparison to obese[10] and normal-weight[11] groups. However, an imaging study in a control group of nonobese individuals,[12] together with research from other groups,[13–15] has shown that the motivation to eat is controlled by an extensive system of reciprocally connected neural areas, beyond the hypothalamus, mediating both the intrinsically derived hunger drive to eat and the extrinsically derived incentive to eat.[12,16] Key brain regions for the former include the hypothalamus[11,17–19] striatum,[20] orbitofrontal cortex (OFC)[21–23] insula[24–26] and anterior cingulate cortex,[24] and, for the latter, the amygdala and OFC.[27–30] Indeed, Lucignani *et al*.[31] have found altered cerebral $GABA_A$ receptor function in the insula and cingulate, frontal and temporal neocortices in six adults with PWS compared to controls. PWS, therefore, provides a unique opportunity to investigate the neural mechanisms linking a syndrome caused by abnormal gene expression to the associated excessive eating behaviour.

We have used positron emission tomography (PET), a whole brain neuroimaging technique, in order to understand how dysfunction in one or more of the aforementioned regions may lead to the severe and lifelong abnormal eating behaviour in PWS. This study has two main elements: first, a comparison of the brain activity associated with fasting and food intake. This examined whether the response of the brain to fasting overnight in those with PWS was similar to those without the syndrome. In addition, based on the above findings by Holland *et al*.,[5] this comparison examined whether a high-energy meal (1200 kcal), the energy value of which was previously found, on average, to be satiating in another PWS group,[5] would be accompanied by a pattern of brain activity similar to that found following food intake in those without the syndrome.[12] Secondly, the brain activity associated with different levels of food intake was examined, in order to compare the activity and associated conscious experience of those with PWS following (a) a meal of equivalent energy value to that previously found to be satiating by those without the syndrome[5] (400 kcal), with (b) the higher-energy meal (as described above – units given in kcal rather than kJ to be consistent with the earlier study).

We predicted that, after an overnight fast, those with PWS would show the same pattern of activation in neural areas associated with hunger as nonobese individuals. Conversely, we predicted an insensitive satiety response, with the pattern of activation in neural areas normally associated with satiety only occurring after consumption of an excessive amount of food.

Methods

Participants

In all, 13 right-handed adults over the age of 21 years with genetically confirmed PWS were recruited from local residential homes and through the Prader–Willi Syndrome Association (UK). Table 1 provides relevant details of those who participated. All those with asthma, diabetes or severe scoliosis were excluded, and any person taking olanzapine was excluded, due to the known effects of weight gain. Of the 13 participants, 12 were living in food-supervised environments, on a daily energy intake that maintained an approximately stable body weight (see Table 1 for details). Written, informed consent was obtained from all of the participants before commencing the study, which was approved by the Cambridge Local Research Ethics Committee and the United Kingdom Administration of Radioactive Substances Advisory Committee.

Experimental protocol

Due to the potential discomfort for the PWS participants, and the difficulties in imaging the OFC using fMRI, PET was chosen. Imaging took place across two consecutive days, in three sessions: after fasting, after a 400 kcal (1674 kJ) breakfast, and after a 1200 kcal (5021 kJ) breakfast. The order of both sessions and trials within each condition were counter-balanced across participants. Participants ate a meal with an energy value of 450 kcal on the evenings before the imaging days at approximately 1700 h and a snack of approximately 50 kcal at approximately 2030 h. The participants had no other opportunity to obtain food and so fasted overnight, equating approximately to a 14-h fast. On the morning of the imaging days, participants were allowed a drink (of negligible calorific value) before leaving for Cambridge, but no food. On arrival at the imaging centre, participants were either given their breakfast or the first imaging session began.

The energy content of each breakfast was based on the results of the previous behavioural study.[5] The energy value of the high-energy meal was equal to, or more than, the whole normal daily energy intake of the participants. The constituents of the breakfasts are shown in Table 2. The breakfasts were designed to look the same, and participants were not informed that they were consuming different

Table 1 Participant details

Participant	*Age*	*M/F*	*IQ*	*BMI*	*Weight (kg)*	*Max. weight (kg)*[a]	*Diet (kcal)*[b]	*FRPQ*[c]	*Medications*
1	31	F	68	32.2	69.85 (stable)	203.21 (21 years)	1200	60	Fluoxetine/Bendrofluazide/Cetirizine
2	42	F	70	35.9	67.58 (stable)	95.25 (20 years)	700–750	66	Procyclidine/Paroxetine/Risperidone/Thyroxine
3	33	F	54	27.8	60.33 (stable+)	130.18 (20 years)	700	54	Trifluoperazine/Procyclidine/Carbamazepine/Sertraline/Haloperidol
4	31	F	—	27.8	62.59 (stable)	82.55 (29 years)	1200	—	Fluoxetine/Flupenthizol/Brevinov/Thyroxine
5	24	M	73	32.4	85.73 (stable−)	165.11 (20 years)	1200	49	Fybogel/Fosamax/Cod Liver Oil
6	22	M	88	30.2	71.7 (stable)	112.94 (19 years)	1200	55	None
7	33	M	73	26.3	76.2 (stable+)	95.25 (16 years)	700–750	50	Sertraline/Risperidone/Fybogel/Docusate Sodium
8	22	F	75	42.6	102.3 (+)	101.6 (22 years)	900–100	28	HRT
9	29	F	76	34	71.21 (stable−)	125.42 (20 years)	1200	54	Micronor
10	23	M	70	29	71.9 (build up)	82.1 (19 years)	1200	66	None
11	27	M	64	36.4	96.16 (stable−)	139.71 (27 years)	1200	39	Andropatch/Docusate Sodium/Sennocide 15/Lansoprazole
12	38	M	73	31.8	78.92 (stable)	133.36 (unknown)	1200	43	Chlorpromazine/Fybogel/Lactulose/Frusemide/Calicipotriol/Lansoprazole
13	26	M	62	22.5	69.09 (stable)	111.13 (24 years)	1200	48	Risperidone
Mean	29	6F/7M	71	31.5	75.7	121.37	1150	51	
s.e.	1.7	—	2.4	1.4	3.4	9.5	—	3.2	

[a]Maximum weight known reported by carer (age at max. in brackets). [b]Energy value of daily food intake. [c]Total score on the Food-related Problems Questionnaire. Key to weight column: stable+, weight stable except home visits (from residential home). stable−, mainly stable but attempting to lose some weight at time of imaging sessions. +gaining weight. Build up – intentionally consuming more calories to reach target weight.

Table 2 Constituents of the 400-kcal and 1200-kcal breakfasts

400-kcal Breakfast			*1200-kcal Breakfast*		
Amount	*Food*	*Energy (kcal)*	*Amount*	*Food*	*Energy (kcal)*
60g	No sugar Alpen[a]	214.2	75 g	Alpen[a]	273.75
150 ml	Semi-skimmed milk	66	150 ml	Full cream milk	99
2.5 g	Sugar	9.85	25 g	Maxijul	Below
0.5 g	Granular Canderel[a]	1.9			
350 ml	No sugar Sunny Delight[a]	35	340 ml	Sainsbury's 100% Squeezed smooth Orange Juice[a]	163.2
110 ml	Total 0% Fat Greek Yoghurt[a]	61.6	110 ml	Total Full Fat Greek Yoghurt[a]	143
35 g	Canned summer fruits in light syrup (drained)[a]	22.75	35 g	Canned summer fruits in light syrup (drained)[a]	22.75
			52 g	Maxijul	Below
0.5 g	Granular Canderel[a]	1.9	132 g	Maxijul[a]	500
	Total	413.2		Total	1201.7

[a]Denotes information from product label. Other figures from McCance and Widdowson's The Composition of Foods. 6th Edition (2002).

energy loads. Each imaging session started approximately 60 min following consumption of the meal, in line with the absorptive phase after a meal when glucose levels are still elevated.[32] In this design, therefore, we were investigating the later stage of satiety, as opposed to the earlier stages of satiation after consumption of food. Visual analogue scales (VAS) were presented prior to the start of, and immediately following, each imaging session.

A blood sample was taken prior to each PET session (as the cannula was inserted) in order to measure plasma levels of glucose, leptin, insulin, ghrelin and PYY. All blood samples were collected on ice and spun within 10 min. Levels of glucose were measured using the hexokinase-glucose-6-phosphate dehydrogenase method. Insulin levels were measured by the 1235 AutoDELFIA automatic immunoassay system using a two-step time-resolved fluorometric assay. For leptin measurements, samples were assayed in duplicate on the AutoDELFIA using a two-step time-resolved fluoroimmunoassay. Plasma was stored at −40°C for radio-immunoassay of ghrelin and PYY. PYY and ghrelin-like immunoreactivity was measured using established radioimmunoassays.[33–36] The ghrelin antibody used in this assay crossreacted with both acylated and des-acylated ghrelin. All samples were assayed in duplicate and within one assay to eliminate interassay variation.

During the separate sessions, each participant received four consecutive scans at 8-min intervals. To keep the cognitive processes constant and focussed on appetitive processes, during each imaging session participants were asked to consider photographs of food (produced by the Foods Standards Agency[37]), and choose the foods they most preferred. The task was based on that used in the control study.[12] The task displays were designed specifically for the study, and were presented on a touch-sensitive screen

controlled by a Pentium microcomputer. Each participant was scanned in the presence of low background noise and dimmed ambient lighting.

An informant-based questionnaire was given to the main carer of each participant as a measure of the range and severity of that participant's food-related problems. The total scores for each participant for the Food-Related Problems Questionnaire[38] (FRPQ) are shown in Table 1.

PET methods

PET procedures were conducted at the Wolfson Brain Imaging Centre (Addenbrooke's Hospital, Cambridge, UK), using the GE Advance System. For each scan, 35 image slices were produced at an intrinsic resolution of approximately $4.0 \times 5.0 \times 4.5\,mm^3$. Each participant received a 20-s intravenous bolus of $H_2{}^{15}O$ through a forearm cannula at a concentration of $300\,MBq\,ml^{-1}$ and a flow rate of $10\,ml\,min^{-1}$. Each scan provides an image of regional cerebral blood flow (rCBF) integrated over a period of 90 s from when the tracer first enters the cerebral circulation.

Data analysis

Images from the three sessions were preprocessed separately for each participant, and then combined for the group statistical analysis, using Statistical Parametric Mapping (SPM99, provided by the Wellcome Department of Imaging Neuroscience, London, UK). Images within each session were realigned to the first image in that session. The images were then reoriented to the anterior commisure in order to covary out movement.[39] This process created movement parameters for each session which were added into the statistical model, as a covariate of no interest, together with a scan time order covariate. The mean realigned images from the second and third imaging sessions were coregistered to the first session, and the means of the three sessions were realigned. Using the mean of the coregistered sessions, the images were normalised for global rCBF value and spatially normalised to the standard brain, based on those from the Montreal Neurological Institute, and, finally, were smoothed using a Gaussian kernel at 12 mm FWHM.

Blood flow changes between experimental conditions were estimated for each voxel according to the general linear model using SPM99. The data for each participant was entered by selecting all 12 images in the order in which they were conducted, and the conditions were then included in the model in order. A number of anatomically specific *a priori* hypotheses, based on PET imaging findings comparing fasting and food intake in a nonobese, non-PWS group,[12] were tested comparing activation patterns in defined subtractions. For the fasting minus 1200 kcal meal contrast, the regions of interest were the hypothalamus, amygdala, striatum, brain stem, insula, and anterior cingulate. For the 1200 kcal meal minus fasting contrast, the regions of interest were the lateral OFC and temporal cortex. An exploratory analysis was conducted to examine the contrasts between the 400- and 1200-kcal meals. The statistical threshold for reporting a peak as significant was set at $P<0.05$, corrected for multiple comparisons.

For the group analysis, participants were divided according to the percentage change in fullness, as rated on the VAS from before imaging in the fasting condition, and before imaging but after consumption of the higher-energy meal (Figure 4a). Six participants were in each group, as one participant (no. 3) from the main group did not complete the 1200 kcal condition. The groups were entered separately into the general linear model, which was otherwise the same as the main analysis.

Data from the VAS and the blood sample measurements were analysed using nonparametric Wilcoxon signed ranks test.

Results

The main comparison described in this section contrasted the fasting condition with the 1200 kcal condition, as this examines the greatest difference and tests the response of the appetitive system in those with PWS to an extreme. The higher-energy meal was most likely to produce satiety in those with PWS, based on previous evidence.[5] The secondary neuroimaging analysis compares the 400 kcal condition with the 1200 kcal condition to assess the brain activity associated with meals of different energy value.

Biochemical data

Mean levels of plasma glucose, insulin, leptin, ghrelin and PYY before and after each imaging session are shown in Table 3. Levels of each measure were significantly different between the fasting and 1200 kcal conditions before the imaging sessions (glucose $Z=-2.805$, $P=0.005$; insulin $Z=-2.366$, $P=0.018$; leptin $Z=-2.380$, $P=0.017$; ghrelin $Z=-2.240$, $P=0.025$; PYY $Z=-2.521$, 0.012). After the imaging sessions, however, levels of insulin ($Z=-2.521$, $P=0.012$), ghrelin ($Z=-2.366$, $P=0.018$) and PYY ($Z=-2.366$, $P=0.018$) were still significantly different between the two conditions, whereas levels of glucose ($Z=-0.981$, $P=0.326$) and leptin ($Z=0.059$, $P=0.953$) were not.

Brain regions associated with fasting and with consumption of the high-energy meal

Our initial analysis was of two main contrasts between the overnight fast and the consumption of the 1200 kcal breakfast. In order to examine the neural substrates of hunger, the relative activation pattern resulting from the fasting condition minus the higher-energy breakfast condition was determined. This contrast revealed relatively greater activation after fasting in those same areas associated with hunger found under similar conditions in nonobese individuals,[12] including the hypothalamus, amygdala, basal ganglia, thalamus and

Table 3 Mean levels (±s.e.) of metabolites and hormones before and after each imaging session

Before/after imaging session	*Fasting condition*		*400-kcal Condition*		*1200-kcal Condition*	
	Before	*After*	*Before*	*After*	*Before*	*After*
Glucose (mmol/l)	4.7±0.18	5.5±1.10	5.61±0.47	5.5±0.55	7.57±0.83	6.79±0.84
Insulin (pmol/l)	22.13±3.11	16.22±2.54	197.88±18.68	159.86±36.98	284.33±46.30	383.44±95.08
Leptin (ng/ml)	29.47±8.37	28.00±7.41	33.29±8.57	34.30±10.72	26.61±7.52	29.33±8.01
Ghrelin (pmol/l)	1364.59±232.42	1222.46±176.30	964.21±106.61	1051.43±154.40	1088.67±138.37	804.74±111.17
PYY (pmol/l)	23.67±3.29	22.324±2.90	31.87±4.22	32.13±3.65	43.77±5.96	33.47±3.73

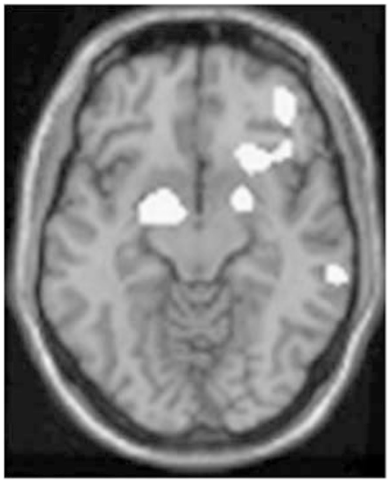

Figure 1 Sagittal section showing significant rCBF changes in the contrast between fasting and the 1200-kcal breakfast. Coordinates of local maxima ($P<0.05$ corrected) are as follows: hypothalamus*,† (−14, −2, −10, $t=6.05$; −2, −2, −2, $t=5.06$), amygdala (14, −6, −32, $t=5.10$), striatum*,† (22, 2, −8, $t=5.23$), thalamus† (−14, −34, 10, $t=5.48$), anterior cingulate† (8, 34, 26, $t=4.17$ activated nonsignificantly at coordinates almost identical to those reported previously), lateral OFC* (42, 46, −6, $t=6.08$), OFC* (area 47/12 m 26, 22, −12, $t=5.00$), inferior temporal cortex* (−12, 8, −36, $t=6.23$; 66, −32, −6, $t=5.83$). * indicates regions shown in the figure and †indicates regions previously reported in nonobese, non-PWS individuals.[12]

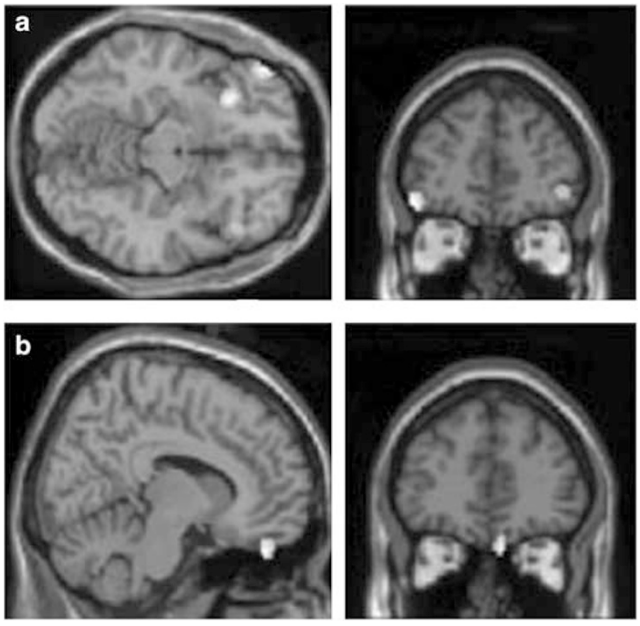

Figure 2 Sagittal and coronal sections showing significant ($P<0.05$ corrected) rCBF changes in the contrast between the 400-kcal (1674 kJ) and 1200-kcal (5021 kJ) breakfasts. (**a**) Illustrates the increased activation in the lateral OFC, bilaterally, following the 400-kcal (1674 kJ) meal. Coordinates in left OFC: −46, 46, −12, $t=7.19$ and −28, 24, −12, $t=6.91$, and right OFC: 46, 50, −6, $t=5.61$ and 46, 28, −12, $t=5.31$. (**b**) Illustrates the increased activation in the medial OFC, following the 1200-kcal meal. Coordinates: 10, 42, −28, $t=4.58$, $P=0.1$ corrected.

anterior cingulate (Figure 1). In addition, the lateral OFC and inferior temporal cortex were activated (Figure 1).

Secondly, to examine the effect of food intake on the pattern of brain activation in PWS, the reverse comparison (higher-energy meal – fasting) was performed. This revealed that, after the higher-energy breakfast, areas of the brain previously shown to be associated with satiation and satiety in non-PWS individuals, including orbitofrontal, temporal and prefrontal cortices,[12,13] did not show increased activity. In fact, no activations survived whole brain correction for multiple comparisons.

Brain regions associated with consumption of the 400- and the 1200-kcal meals

A further analysis was conducted in order to contrast the neural activity following the breakfast of 400 kcal (1674 kJ) with that of the higher-energy breakfast (1200 kcal, 5021 kJ). Consumption of the 400 kcal (1674 kJ) meal resulted in greater activation in lateral OFC, bilaterally (Figure 2a), including a peak at a similar location to that found after fasting. In contrast, consumption of a meal comprising three times the energy resulted in activation of the medial OFC, although marginally missing significance according to our conservative criteria (Figure 2b).

Analysis of subjective ratings of hunger, fullness and desire to eat

To assess the subjective experience that accompanied the states captured by imaging, ratings from visual analogue scales measuring hunger, fullness and desire to eat given before and after each imaging session were examined. Ratings before the imaging sessions were significantly different in the expected direction when the fasting and

higher-energy conditions were compared (hunger $Z=-2.906$, $P=0.004$; fullness $Z=-2.936$, $P=0.003$; desire to eat $Z=-2.756$, $P=0.006$; Figure 3a). However, immediately after the imaging sessions, even after consumption of the higher-energy meal only 2 h before, the ratings had already returned to levels that were not significantly different from ratings obtained after fasting overnight (hunger $Z=-1.726$, $P=0.084$; fullness $Z=-1.869$, $P=0.062$; desire to eat $Z=-0.657$, $P=0.511$; Figure 3b).

Percentage change in fullness ratings relates to orbitofrontal activity

Given the lack of activity in satiety-related areas in the comparison between the higher-energy meal and fasting conditions (see above), *post hoc* analysis of the extent of change in fullness ratings from the fasting baseline to after consumption of the higher-energy meal was performed. This indicated that only half the participants showed a substantial shift in their fullness ratings (Figure 4a). *Post hoc* analysis of the imaging data, split according to the percentage change in fullness after food, showed a clear response in the medial OFC ($x=10$, $y=44$, $Z=-28$) following the higher-energy meal in the group whose ratings shifted to fullness ($t=6.71$, $P<0.0001$ corrected, Figure 4b), but not in the group whose ratings did not shift ($t=0.73$, $P=1.0$ corrected; local maxima $t=0.80$). The fact that a medial OFC response was clearly absent in half of the participants also accounts for why activity in this region just missed statistical significance in the combined group analysis described above.

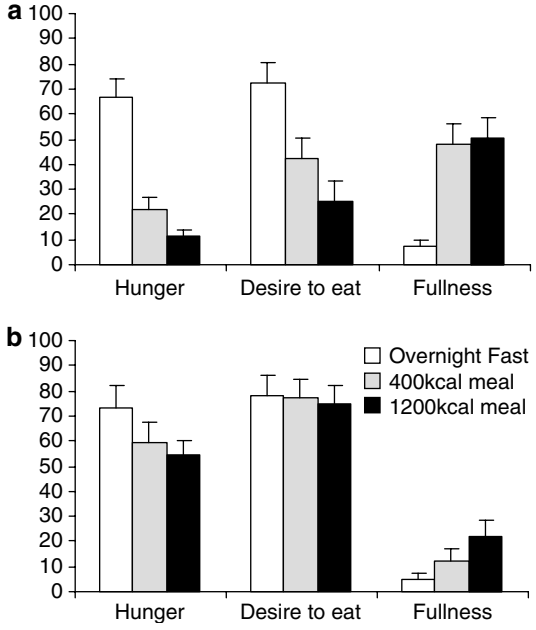

Figure 3 Mean (±standard error) ratings on visual analogue scales. (**a**) Before each imaging session for questions: how hungry do you feel? how strong is your desire to eat now? and how full do you feel? (**b**) After each imaging session.

Further analyses were conducted to discover whether there were any differences between the two groups that could explain the differential brain activity. No systematic differences between the groups were found according to gender, age, IQ, BMI, maximum weight ever, weight lost at scan time since maximum, total daily intake of energy or medications ($P>0.05$). In addition, no difference between the groups was found according to the total or individual subscale scores of the FRPQ.

Discussion

The findings from this study demonstrate that there is an abnormal neural response to the food intake in those with PWS. After fasting overnight, brain regions previously shown to be associated with hunger[12] showed a relative increase in activation, compared to the high-energy meal. In addition, other areas that have been associated with the anticipation of food when hungry were activated.[23,26,40] Exposure to food stimuli may lead to greater anticipation of future food intake in those with PWS than in those without. Notably, we found that the normal neural response to food intake was absent, as the high-energy meal did not result in a pattern of neural activity similar to that seen in those without PWS after food intake. We had hypothesized that the PWS genotype would result in a delayed response to food intake and that a larger meal would have resulted in a pattern of cerebral activation characteristic of satiety. Support for this hypothesis was not

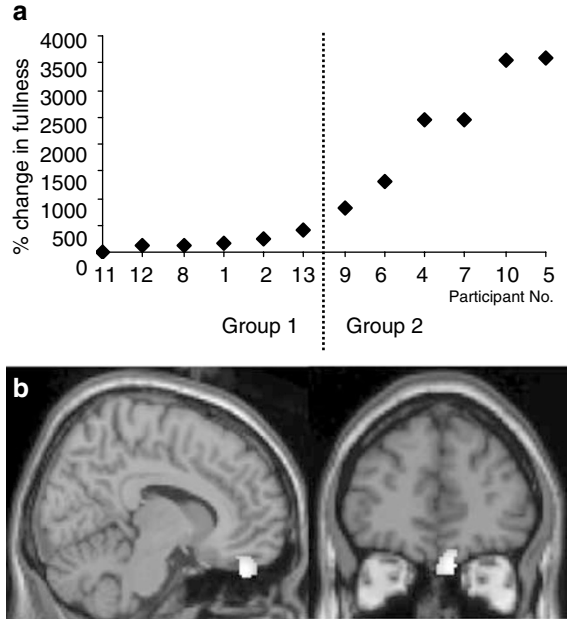

Figure 4 (**a**) Percentage shift in fullness ratings from fasting baseline to after 1200-kcal (5021 kJ) meal. The group was divided into two for further *post hoc* analysis of the imaging data. (**b**) Sagittal and coronal sections illustrate the significant medial OFC activity in the group (2) whose ratings did shift to fullness (10, 44, −28, $t=6.71$, $P<0.0001$, corrected).

found, however, even after consumption of a meal previously found to be satiating by a group of people with PWS,[5] which was equal to or more than the whole normal daily energy intake of the participants in this study.

In contrast to the imaging results, the blood plasma levels of glucose, insulin, ghrelin and PYY were found in accordance with the predictions: levels of all of the above were significantly different between the fasting and high-energy meal conditions. While ghrelin levels were elevated in comparison to controls in previously published literature,[35,41–43] recent work has shown that even when ghrelin levels are reduced to a normal level, using somatostatin or a somatostatin agonist, there is no effect on appetite or food intake in individuals with PWS.[44,45] This suggests that the dysfunction in PWS is central in origin, rather than as a result of abnormalities in the periphery, at least in terms of the measures in this study. Analysis of the subjective ratings showed that hunger and desire to eat decreased as the energy value of food consumed increased, but returned to fasting levels only approximately 2 h after eating the high-energy meal. While the ratings of fullness were somewhat reduced after food intake, they were not significantly different following the two meals, and, again, showed a rapid shift back to fasting levels. This provides further evidence that it is the process of satiety and/or perception of that state that is abnormal in PWS.

Given some change in the mean fullness ratings after food intake was found, further analysis was warranted to examine whether patterns of activation in the comparison between the two meals might account for the fact that some people with PWS had reported changes in ratings as would be expected after significant food intake. On comparison of the activation following consumption of the 400- and 1200-kcal meals, a dissociation was found in the OFC; lateral OFC activity was associated with the 400-kcal meal, whereas medial OFC activity was associated with 1200-kcal meal. Converging evidence indicates that the functions of these regions can be dissociated; the medial OFC has been shown to monitor reward value, whereas the lateral OFC evaluates punishment value, which may lead to an adjustment of current behaviour.[46] Given this dissociation, we suggest that the contrasting orbitofrontal activations indicate that a meal of controlled energy value, but still meeting the normal energy intake for breakfast (400 kcal), leaves the person with PWS unsatisfied and wanting more, which may at some level be a distressing experience that, together with the absence of a satiety response in other cortical areas, maintains the drive to eat. However, consumption of a meal of greater energy value, even without knowledge of the true content, leads, at least temporarily, to a more satisfied and pleasurable response that we hypothesize may then be perceived by those with PWS as a change in hunger and fullness.

Further analysis of the percentage change in fullness supported our hypothesis that, for some those with PWS, it is the change in activation from the lateral to the medial OFC that corresponds to a shift in hunger and fullness ratings, leading to a positive emotional state. Recent evidence does suggest that the OFC is a key brain region involved in linking food with hedonic experience.[15] Moreover, it was interesting to find that only half the group showed this medial OFC activity following the 1200-kcal meal. No differences were found, however, between the group who did show this activity and who showed a large shift in their percentage fullness and those who did not, in terms of demographic and anthropometric characteristics and medications. It was only in contrast with the lower-energy meal that the medial OFC activity was seen in association with the high-energy meal for some participants. In comparison to the 1200-kcal meal, the lower-energy meal could be argued to induce a more unsatisfying experience than the fasting condition, as a small amount of food may serve to increase appetite further.

The main finding from this study, of an insensitive satiety response in PWS, supports preliminary findings from another functional neuroimaging study in PWS.[9] The results of this study extend these findings, first, by examining a later postprandial time frame, and, secondly, by showing that even a high-energy meal did not result in a pattern of neural activity similar to that seen in those without PWS after food intake.[12] In this study, we have taken a behavioural hypothesis-driven approach to the design, whereby we have used fixed-energy meals, based on the findings of the aforementioned eating behaviour study in PWS,[5] to investigate the neural basis of hunger and satiety in those with this syndrome. Different experimental approaches have been used to examine similar questions; some designs have involved feeding a liquid meal (a proportion of the individual's total energy requirements) to participants while in the scanner,[24] or giving participants a fixed-energy glucose drink.[9,11] The aim of this study was to measure changes in neural activity and the conscious experience of the participants with PWS after consumption of a normal meal, increasing the ecological validity of the design. It is acknowledged that using fixed-energy meals does not take individual energy requirements into account; however, for the higher-energy meal at least, some participants consumed a significant proportion of their total daily energy requirement – yet a pattern of neural activation representative of satiety was still not found.

The above discussion of the findings of this study should be considered in the light of the limitations of the study. It is noted that a number of the participants with PWS were on medications that may have had a confounding effect on the rCBF found in association with fasting and food intake, and/or the perception of satiety. While it is recognised that a medication-free sample would have been ideal, PWS is a rare syndrome associated with many health problems.[47] Importantly, none of the medications had been prescribed to alleviate the eating problems and all of the participants were still demonstrating eating problems despite the medications, as indicated by scores on the FRPQ. A potential limitation of this study design is that there is no direct control group who

consumed the 400- and 1200-kcal meals. This comparison, however, would not be equivalent, and as such, as meaningful or relevant, in those without the syndrome, because in a non-PWS group, it would be expected that the lower-energy meal may result in a pattern of neural activation associated with satiation and satiety, but that the higher-energy meal would take them beyond a pleasant, normal state of satiety, resulting in different brain activation accordingly. Finally, it should be noted in the interpretation of these results that PWS is associated with learning disability. It is not clear, however, how this learning disability could produce such a pattern of results as reported here, particularly with consideration for the specific hypotheses and results from the non-PWS study.[12]

In conclusion, the results of this study, with consideration for the limitations, suggest that the neural representation of hunger and associated experience is similar in those with PWS to those without the syndrome. On the basis of the imaging and behavioural data, it appears that there is a dysfunction in the satiety system in those with PWS, which may be the basis of the hyperphagia seen in those with the syndrome. When comparing the neural activity following meals of different energy values, the resulting activation of the medial and lateral OFC suggests that some of those with PWS can detect a change in their internal states, which they interpret as increasing fullness. However, the impaired neural response to a higher-energy meal suggests that they do not experience fullness in the same way as those without the syndrome. These findings provide evidence for a link between the underlying genetics of PWS, the dysfunctional processing of neural satiety systems and the abnormal eating behaviour.

The main implication of these findings for the management of PWS is that the supervision of the food environment is critically important at all times to avoid severe obesity and associated health problems. Consideration should also be given to the genetic basis of this syndrome and the possible implications of these findings to the general population. While research into the neural basis of satiety in PWS is in its infancy, the findings from this study imply that, for the neural satiety system to function, involving OFC, temporal and prefrontal cortices, the normal expression of the paternal allele of the maternally imprinted 'PWS' gene(s) may be required. Identifying this gene(s) in the PWS critical region associated with the abnormal eating behaviour, its modes of action and sites of expression will be significant for people with PWS and possibly for understanding obesity in the general population.

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