Calorie Anticipation Alters Food Intake After Low-Caloric but Not High-Caloric Preloads

P.S. Hogenkamp¹, J. Cedernaes¹, C.D. Chapman¹, H. Vogel², O.C. Hjorth¹, S. Zarei¹, L.S. Lundberg¹, S.J. Brooks¹, S.L. Dickson², C. Benedict¹ and H.B. Schiødth¹

Objective: Cognitive factors and anticipation are known to influence food intake. The current study examined the effect of anticipation and actual consumption of food on hormone (ghrelin, cortisol, and insulin) and glucose levels, appetite and *ad libitum* intake, to assess whether changes in hormone levels might explain the predicted differences in subsequent food intake.

Design and Methods: During four breakfast sessions, participants consumed a yogurt preload that was either low caloric (LC: 180 kcal/300 g) or high caloric (HC: 530 kcal/300 g) and was provided with either consistent or inconsistent calorie information (i.e., stating the caloric content of the preload was low or high). Appetite ratings and hormone and glucose levels were measured at baseline (*t* = 0), after providing the calorie information about the preload (*t* = 20), after consumption of the preload (*t* = 40), and just before *ad libitum* intake (*t* = 60).

Results: *Ad libitum* intake was lower after HC preloads (as compared to LC preloads; *P* < 0.01). Intake after LC preloads was higher when provided with (consistent) LC information (467 ± 254 kcal) as compared to (inconsistent) HC information (346 ± 210 kcal), but intake after the HC preloads did not depend on the information provided (LC information: 290 ± 178 kcal, HC information: 333 ± 179 kcal; caloric load*information* *P* = 0.03). Hormone levels did not respond in an anticipatory manner, and the post-prandial responses depended on actual calories consumed.

Conclusions: These results suggest that both cognitive and physiological information determine food intake. When actual caloric intake was sufficient to produce physiological satiety, cognitive factors played no role; however, when physiological satiety was limited, cognitively induced satiety reduced intake to comparable levels.

*Obesity* (2013) 21, 1548-1553. doi:10.1002/oby.20293

Introduction

Cognitive factors are known to influence short-term food intake in humans (1,2): food anticipation as well as expectations regarding energy content, macronutrient content, or health aspects of food have been shown to influence intake (3–6), taste perception and preference (7,8), and expected and perceived satiety (9). Moreover, food anticipation affects physiological responses involved in the control of food intake. For example, external cues, such as food pictures (10) or labels providing “indulgent” information (11), elevate levels of the orexigenic hormone ghrelin. Ghrelin plays an important role in the short-term control of food intake: it stimulates meal initiation and produces a quick and robust increase in consumption (12,13). Ghrelin concentrations rise in humans during meal anticipation (14) and decrease in proportion to the amount of calories consumed (15,16). In a recent study, postprandial suppression of ghrelin levels was stronger in individuals who anticipated food intake as compared to those who did not expect a meal (17). Also, HPA-axis activity may be affected by food anticipation (18), therewith possibly modulating cortisol levels that are observed after food intake (19–21). However, it is not clear whether cognitive cues modulate (postprandial) ghrelin responses when the anticipated foods differ in their energy content and whether...
these changes in hormone levels may contribute to possible differences in subsequent food intake. To test this, participants in the present study were offered two low-caloric (LC) and two high-caloric (HC) preloads over four visits. Both the LC and HC preloads were presented once with consistent calorie information (i.e., low or high) and once with inconsistent calorie information (i.e., high or low). We assessed pre-prandial and post-prandial plasma levels of ghrelin, as well as cortisol concentrations. In addition, LC foods (grapes), HC foods (muffins), and foods with an intermediate energy density (bread) were offered \textit{ad libitum} 30 min following the preload. Energy intake was calculated to determine whether cognitive factors modulate subsequent food intake. We hypothesized that \textit{ad libitum} food intake would be lower following the preloads that were presented with LC information and that incongruent calorie information would modulate the appetite hormone responses (ghrelin, insulin, and cortisol).

**Methods**

**Participants**

In a randomized, cross-over design with four conditions, 12 healthy young women [age: $23.7 \pm 1.8$ y, BMI: $22.8 \pm 1.9$ kg/m$^2$], were enrolled. They had no history of eating disorders, no dietary restrictions, and were not on any medications. Restraint scores were calculated to determine whether participants had a restrained eating pattern. Participants were included if they were in the age range of 20-25 years, had a BMI between 18.5 and 25 kg/m$^2$, and were not taking any medication. Participants were also asked to maintain their normal dietary intake and physical activity levels during the study period. Written informed consent was obtained from all participants before the start of the study, and the study was approved by the regional ethics committee of Uppsala (EPN), and the study is registered with ClinicalTrials.gov (NCT01680315).

**Design and procedures**

Participants were instructed to refrain from alcohol, caffeine, food intake, and drinks except water after 22.00 the day before each test day and not to consume anything in the morning before arrival at the research centre at 07.30. Each session took place on a separate testing day, with a minimum wash-out period of 5 days between each session. Blood was sampled four times, at 0, 20, 40, and 60 min after inserting and using a venous cannula. Immediately after each blood sample, participants rated their appetite sensations (hunger, fullness, desire to eat, and prospective consumption) and thirst on 100-mm visual analogue scales (VAS), anchored “not at all” and “extremely”. Product information (described below) was provided after the first blood sample ($t = 0$), participants tasted, rated, and consumed the preload after the second blood sample ($t = 20$), and after the third blood sample ($t = 40$), participants filled out an evaluation questionnaire. Finally, they received their \textit{ad libitum} breakfast after the fourth blood sample ($t = 60$). Following \textit{ad libitum} consumption, participants once more evaluated their appetite sensations ($t = 80$; Figure 1). For tasting and rating the preloads, participants consumed one spoonful and evaluated the perceived pleasantness, sweetness, sourness, creaminess, and thickness of the yogurt on a 100-mm VAS, anchored “not at all” and “extremely”. This was repeated in the evaluation questionnaire. This procedure was repeated for all four conditions. The order of conditions was randomized within and balanced between participants.

**Test foods**

Participants were served 300 g of LC yogurt (60 kcal/100 g) or HC yogurt (177 kcal/100 g) preloads. The HC preload consisted of 219 g of commercially available mild natural yogurt (Mild lätthygurtt naturell, Arla Foods), 66 g of table sugar, and 15 g of sunflower oil (Zeta). To match the LC preload in sweetness, 1.92 g artificial sweetener (cyclamate; Suketter; Cederroth International) dissolved in 8 g of water was added to 290 of the yogurt. The preloads were served in transparent cups.

![FIGURE 1 Experimental design. Blood was sampled every 20 min, and participants evaluated their appetite sensation (hunger, fullness, desire to eat, prospective consumption, and thirst) on a 100-mm visual analogue scale immediately after each blood sample.](#)
After the preload, participants were provided with a breakfast plate that consisted of 300 g of white grapes (64 kcal/100 g), 10 triangles of fruit bread (~18 g each) with 8 g of butter (312 kcal/100g; Frukt–kusaar; Fazer), and 5 muffins of 20 g each (430 kcal/100g; Citron–muffins; Hägges). Participants were provided an additional plate to avoid “empty your plate” behavior (23,24). This happened only twice, with the same participant, and both times when the provided product information stated LC content.

Calorie information
In the “low-calorie information”-condition (LC-info), we provided a product description that stated that the preload is “a healthy breakfast product,” “light and fresh,” “low in fat and calories,” and “without added sugar”. We also provided a list of ingredients (including nonfat milk and vitamin A and D) and nutrition facts [energy (kcal) and macronutrients (g/100 g)]. In addition, participants were asked to calculate the energy percentage of protein, the total amount of calories in the product, and to describe the characteristics (picture, color, etc.) of a label and taste that would go best with the product. Every time the word “food” or “yogurt” was mentioned, it was preceded by the adjective LC. In the “high-calorie information”-condition (HC-info), the questionnaire was the same except for the product information (i.e., “a breakfast treat with the finest ingredients”, “that keeps away hunger flaws during the morning”, “with creamy yogurt cultured from whole milk”, and “rich product”), ingredient list (whole milk and milk proteins), nutrition facts, and the HC adjective.

When participants received the specific calorie information for the second time, they were asked to carefully consider the questionnaire. In addition to the previous visit with the same calorie information, participants also calculated the contribution of the preload to their daily energy requirements and compared the characteristics of the label (that they described) with food labels used for commercially available LC (HC) dairy products.

Biochemical analysis
Blood samples were centrifuged directly after sampling, and the supernatant was stored at −80°C, for analysis of plasma glucose, insulin, cortisol, and ghrelin. Plasma glucose was measured using routine assays (hexokinase method, Aeroset; Abbott Diagnostics, North Chicago, IL). Noncompetitive immunometric assays were used to determine serum concentrations of insulin (12017547 122; Roche Diagnostics, Mannheim), and competitive assays were used to determine cortisol concentrations (11875116 122; Roche Diagnostics). Total ghrelin concentrations were assessed using commercially available ELISA kits for humans (EZGRT-89K; Millipore, Billerica, MA). Levels of total ghrelin were out of range for six samples of one participant. We therefore excluded all data from this participant for the ghrelin analyses.

Data analysis
Continuous variables are presented as means (± SD). ANOVA (repeated measures) was used to test the effects of caloric content (content) and/or calorie information (info) on ad libitum energy intake, appetite ratings, blood parameters, and sensory attributes. Concentrations of blood parameters at $t = 0$ were included as a covariate, to adjust for differences across conditions. Tukey’s post hoc tests were used to test for ANOVA-indicated differences, between the conditions. We included the time variable in the model to test for a time effect for the appetite ratings and blood parameters, and we tested for the effect of caloric content and caloric information on appetite sensations and blood parameters at the individual time points. Data were analyzed using SAS (version 9.3; SAS Institute). Results at a $P$-value of <0.05 were considered significantly different.

Results
Ad libitum energy intake
The amount of energy consumed from the breakfast plate depended on the caloric content of the preloads ($P < 0.01$) and on the information provided (content*info interaction: $P = 0.03$): ad libitum intake was higher when the LC preload was provided with the LC-info (Figure 2). Including BMI, body weight and/or restraint scores as covariates in the model did not change the results. Only two participants consumed at least one muffin. Repeating the analyses without the muffins, that is, including grapes and fruit buns only, did not change the result patterns.

Blood parameters
Baseline concentrations ($t = 0$) of glucose, insulin, and cortisol did not differ across conditions, but we observed higher ghrelin concentrations in the HC-info conditions at baseline ($P = 0.03$) (Figure 3). Both glucose and insulin levels depended on the caloric content (main effect content: glucose $P = 0.01$; insulin $P < 0.001$), with higher concentrations after consuming the HC preloads as compared to the LC preloads ($t = 40$ and $t = 60$). Cortisol concentrations depended on the provided caloric information (main effect: $P = 0.05$), but post hoc tests did not indicate significant differences between conditions, and cortisol concentrations did not differ across conditions at individual time points. Total ghrelin concentrations depended on the caloric content (main effect: $P < 0.01$), but post hoc tests did not show significant differences between conditions (Figure 3).
Appetite ratings
As expected, hunger, desire to eat, and prospective consumption decreased, and fullness increased over time (all $P < 0.0001$). Ratings before ($t = 0$ and $t = 20$) and immediately after consuming the preload ($t = 40$) did not differ across conditions for any of the appetite sensations. At $t = 60$, we observed an interaction effect for both hunger and fullness (content*info: $P < 0.01$), while main effects of energy content and information were not significant. Post hoc tests showed that participants reported to be more hungry and after consuming the HC preload provided with the HC-info condition as compared to the HC preload with the LC-info ($P = 0.02$) but did not indicate significant differences for fullness (Figure 4). Appetite sensations did not differ across conditions after ad libitum intake of items at the breakfast plate ($t = 80$).

Sensory attributes and pleasantness
We did not observe differences in pleasantness ratings of the different preloads. The LC and HC preloads (independent of the information provided) were considered to be equally sweet and sour.
Participants perceived the first bite of the LC preloads as more creamy ($P = 0.09$) and thicker ($P = 0.06$), but neither of these differences reached significance in the current sample (Table 1). However, ratings following consumption of the preload in its entirety showed the same pattern and reached significance (effect caloric content: creaminess $P = 0.04$; thickness $P = 0.02$). None of the participants guessed the exact aim of the experiment; three of them suggested that the aim was to study the effect of sweeteners and sugars on physiological responses.

### Discussion

The current study aimed to examine whether a mismatch between caloric content and caloric content label information of a preload determines subsequent ad libitum food intake and circulating appetite hormone levels (ghrelin, insulin, and cortisol) in healthy normal-weight women. To this aim, 12 female university students participated in four separate conditions: in two conditions, they consumed a LC preload (180 kcal), and in the other two conditions they consumed a HC preload (530 kcal). In addition, before consumption of the preload in its entirety, they were provided with the calorie content information that a preload is energy dense represents a sufficient stimulus to reduce subsequent ad libitum food intake in young female adults. In line with our findings, previous studies have demonstrated that having the belief that one has eaten a considerable amount of calories influences fullness and feelings of satiety (28,29). However, at this point, it is important to emphasize that the inconsistent caloric content information only affected subsequent food intake in the LC preload conditions, that is, no such effects were seen for the HC conditions. One explanation for these discrepant results might be that once the preload has supplied a certain number of calories, cognitive processes related to food intake play an inferior role for subsequent eating.

There are some mechanisms through which the inconsistent caloric content information in the LC conditions may have reduced subsequent ad libitum food intake in our study. For instance, assuming that the preload was dense in energy may have resulted in increased activation of neural circuits involved in suppression of hunger and food intake, such as the dorsolateral prefrontal cortex (30). It has also been observed that labeling food as low fat can increase intake (31). However, this discrepancy is most likely caused by differences in study settings: meal anticipation compared to no meal anticipation in all conditions, as well as differences in the studied time period (11). Limitations in our study may also have led to discrepancies from prior results. First, we did not select participants based on a restrained eating behavior score. A high dietary restraint score refers to the tendency to control

| TABLE 1 Ratings on pleasantness and sensory attributes (mean ± SD) for the low-caloric (LC) and high-caloric (HC) preloads (irrespective of the caloric information) and for the preloads provided with the LC-info and HC-info (irrespective of caloric content), as well as ratings of the four preloads |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
|                 | LC foods        | HC foods        | Low info        | High info       | LC info          | HC info          | LC info          | HC info          |
| Pleasant        | 50 ± 27         | 44 ± 28         | 45 ± 29         | 49 ± 27         | 46 ± 30          | 54 ± 25          | 44 ± 29          | 45 ± 28          |
| Sweet           | 84 ± 16         | 76 ± 21         | 82 ± 16         | 78 ± 21         | 83 ± 20          | 86 ± 11          | 82 ± 11          | 70 ± 26          |
| Sour            | 19 ± 21         | 16 ± 16         | 17 ± 19         | 18 ± 18         | 22 ± 24          | 15 ± 17          | 11 ± 12          | 20 ± 19          |
| Creamy          | 63 ± 24         | 51 ± 24         | 52 ± 25         | 61 ± 22         | 61 ± 24          | 65 ± 24          | 44 ± 25          | 58 ± 21          |
| Thick           | 53 ± 19         | 41 ± 23         | 45 ± 22         | 50 ± 21         | 53 ± 20          | 54 ± 19          | 36 ± 21          | 47 ± 24          |

$^{a}$LC preloads were perceived more creamy ($P = 0.09$) and thicker ($P = 0.06$).
food intake at a cognitive level (33). Including only restrained eaters may have resulted in a more pronounced effect of the calorie information on food intake. Second, we had a relatively low blood sampling frequency, which may have masked possible alterations of the measured hormones. Finally, we only studied normal-weight healthy females and generalization to males or other weight abnormalities is not appropriate. In summary, our results suggest that both metabolic and cognitive features of food affect subsequent food intake decisions in healthy young women. Bearing the small sample size in mind, additional studies are needed to elucidate further the effects and underlying mechanisms through which cognitive cues related to food intake affect subsequent eating behavior in humans.

Acknowledgments

We thank all participants, Jenny Högblad, Linda Årström, Louise Nilsson, and Monika Gelotte, for their help in carrying out the study. PSH, CDC, OH, SJB, CB, and BHS designed the study, PHS and OH wrote the protocol, PSH, JC, HV, SZ, LSL, and SLD collected the data and conducted analyses; all authors contributed to and have approved the final manuscript.

© 2013 The Obesity Society

References

1. Blundell JE. The control of appetite: basic concepts and practical implications. Schweiz Med Wochenchr 1999;129:182–188.
2. Herman CP, Polivy J. Normative influences on food intake. Physiol Behav 2005;86:762–772.
3. Shide DJ, Rolls BJ. Information about the fat content of preloads influences energy intake in healthy women. J Am Diet Assoc 1995;95:993–998.
4. Wooley SC. Physiologic versus cognitive factors in short term food regulation in the obese and nonobese. Psychosom Med 1972;34:62–68.
5. Caputo FA, Mattes RD. Human dietary responses to perceived manipulation of fat content in a midday meal. Int J Obes Relat Metab Disord 1993;17:237–240.
6. Provencher V, Polivy J, Herman CP. Perceived healthiness of food. If it’s healthy, you can eat more! Appetite 2009;52:340–344.
7. Yeomans MR, Chambers L, Blumenthal H, et al. The role of expectancy in sensory and hedonic evaluation: The case of smoked salmon ice-cream. Food Qual Prefer 2008;19:565–573.
8. Gould NJ, Zandstra EH, Yeomans MR. Manipulating expectations about the calorie content of a breakfast influences acquired liking. Appetite 2011;57:558–558.
9. Fay SH, Hinton EC, Rogers PJ, et al. Product labelling can confer sustained increases in expected and actual satiety, in British Feeding and Drinking Group. Belfast, 2011.
10. Schüessler P, Klagge M, Yassouridis A, et al. Ghrelin levels increase after pictures showing food. Obesity 2012;20:1212–1217.
11. Crum AJ, Corbin WR, Brownell KD, et al. Mind over milkshakes: mindsets, not just nutrients, determine ghrelin response. Health Psychol 2011;30:424–429.
12. Cummings DE, Purnell QJ, Frayo RS, et al. A preprandial rise in plasma ghrelin levels suggests a role in meal initiation in humans. Diabetes 2001;50:1714–1719.
13. Druce MR, Wren AM, Park AJ, et al. Ghrelin increases food intake in obese as well as lean subjects. Int J Obes Relat Metab Disord 2005;29:1130–1136.
14. Cummings DE, Frayo RS, Marmonier C, et al. Plasma ghrelin levels and hunger scores in humans initiating meals voluntarily without time- and food-related cues. Am J Physiol Endocrinol Metab 2004;287:E297–E304.
15. Callahan HS, Cummings DE, Pepe MS, et al. Postprandial suppression of plasma ghrelin level is proportional to ingested caloric load but does not predict intermeal interval in humans. J Clin Endocrinol Metab 2004;89:1319–1324.
16. Le Roux CW, Patterson M, Vincent RP, et al. Postprandial plasma ghrelin is suppressed proportional to meal calorie content in normal weight but not obese subjects. J Clin Endocrinol Metab 2005;90:1068–1071.
17. Ott V, Friedrich M, Zemlin J, et al. Meal anticipation potentiates postprandial ghrelin suppression in humans. Psychoneuroendocrinology 2012;37:1096–1100.
18. Ott V, Friedrich M, Pelip S, et al. Food anticipation and subsequent food withdrawal increase serum cortisol in healthy men. Physiol Behav 2011;103:594–599.
19. Holl R, Fehm HM, Voigt KH, et al. The ‘midday surge’ in plasma cortisol induced by mental stress. Horm Metab Res 1984;16:158–159.
20. Feillet CA. Food for thoughts: feeding time and hormonal secretion. J Neuroendocrinol 2010;22:620–628.
21. Martens EA, Lemmens SG, Adam TC, et al. Sex differences in HPA axis activity in response to a meal. Physiol Behav 2012;106:272–277.
22. Cappelleri JC, Bushmalin AG, Gerber RA, et al. Psychometric analysis of the Three-Factor Eating Questionnaire-R21: results from a large diverse sample of obese and non-obese participants. Int J Obes 2009;33:611–620.
23. Birch LL, McPhee L, Shoba BC, et al. “Clean up your plate”: effects of child feeding practices on the conditioning of meal size. Learn Motiv 1987;18:301–317.
24. Wansink B, Sobal J. Mindless eating: the 200 daily food decisions we overlook. Environ Behav 2007;39:106–123.
25. Higgs S. Memory for recent eating and its influence on subsequent food intake. Appetite 2002;39:159–166.
26. Morewedge CK, Huh YE, Vosgerau J. Thought for food: imagined consumption reduces actual consumption. Science 2010;330:1530–1533.
27. Higgs S, Robinson E, Lee M. Learning and memory processes and their role in eating: implications for limiting food intake in overeaters. Curr Obes Rep 2012;1:91–98.
28. Brunstrom JM, Brown S, Hinton EC, et al. “Expected satiety” changes hunger and fullness in the inter-meal interval. Appetite 2011;56:310–315.
29. Wooley OW, Wooley SC, Dunham RB. Can calories be perceived and do they affect hunger in obese and nonobese humans? J Comp Physiol Psychol 1972;80:250–258.
30. Van den Eynde F, Koskina A, Syrad H, et al. Repetitive transcranial magnetic stimulation reduces cue-induced food craving in bulimic disorders. Biol Psychiatry 2010;67:793–795.
31. Wansink B, Chandon P. Can “low-fat” nutrition labels lead to obesity? J Mark Res 2006;43:605–617.