Original Research Communications

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Long-term habituation to food in obese and nonobese women¹⁻³

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ABSTRACT

Background: Habituation is a form of learning in which repeated exposure to a stimulus leads to a decrease in responding. Eating involves repeated presentation of the same food stimulus in a meal, and habituation is reliably observed within a meal such that faster rates of habituation are associated with less energy intake. It is possible that repeated presentation of the same food over days will lead to long-term habituation, such that subjects habituate to foods repeated over meals. However, no research on long-term habituation to food in humans has been conducted.

Objective: The current study was designed to assess long-term habituation in 16 obese and 16 nonobese premenopausal women.

Design: Obese and nonobese women (aged 20–50 y) were randomly assigned to receive a macaroni and cheese meal presented 5 times, either daily for 1 wk or once per week for 5 wk.

Results: In both obese and nonobese women, daily presentation of food resulted in faster habituation and less energy intake than did once-weekly presentation of food.

Conclusions: Long-term habituation was observed when the same food was presented at daily meals but not when presented once weekly for 5 wk. These results provide the first evidence of long-term habituation to food in women and show that memory of food over daily meals can increase the rate of habituation and reduce energy intake. This trial was registered at clinicaltrials.gov as NCT01208870. *Am J Clin Nutr* 2011;94:371–6.

INTRODUCTION

Habituation is a form of learning in which a decrement in behavioral and physiologic responses to a stimulus is caused by repeated presentation of a stimulus, which does not involve sensory adaptation/fatigue or motor fatigue. People habituate to repeated presentations of the same food within a meal (1). The presentation of a novel food causes dishabituation (2), and stimulus specificity of habituation is observed for the habituating food (3). The rate of habituation to food is related to food consumption, because slower rates of habituation are associated with greater energy intake (4–6). The rate of habituation within an eating session differs between obese and lean subjects, with obese subjects habituation also predicts weight gain in children, because those who habituate slower show greater gains in BMI z scores than do those who habituate at a faster rate (9).

In addition to repeated stimuli having short-term effects on responses, long-term habituation may be observed so that stimulus presentations influence responses over longer intervals. For example, repeatedly presenting the same auditory stimulus within 5 daily sessions resulted in short-term habituation during each session and long-term habituation over the 5 d (10). Similarly, long-term habituation of objective and subjective measures or sexual arousal was observed for participants provided the same erotic stimuli at weekly intervals, whereas no habituation was observed over the same interval if the erotic stimuli were varied (11). In theory, long-term habituation could be observed over multiple meals, such that if a food is presented repeatedly over meals or days, long-term habituation may be observed, which leads to a reduction in energy intake (1).

No research on long-term habituation to food in humans has been conducted, but it is to be expected that the interval between presentations of the same food would influence the process of long-term habituation. Repeated presentations of the same food within a short time interval should be expected to lead to habituation over days, such that the person responds less to obtain the repeated meal than to the first meal. However, longer intervals may diminish the effects of previous food presentations; thus, if sufficient time elapses between presentations, then spontaneous recovery may occur (12) and long-term habituation may not be observed.

Obese adults (7, 13) or overweight children (5, 8) habituate at slower rates to repeated food stimuli than do leaner subjects.

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Given that short-term habituation during a meal is slower for obese subjects, it is possible that obese subjects would be less affected by long-term habituation across meals than would leaner subjects. The current study had 2 primary aims. First, the effects of 5 daily compared with weekly presentations of the same food on habituation were assessed. Second, the influence of obesity on long-term habituation was tested.

SUBJECTS AND METHODS

Participants

The participants were 16 nonobese and 16 obese females between 20 and 50 y of age, who were recruited from flyers and a preexisting database. The participants were considered nonobese if their body mass index (BMI; in kg/m²) was <30 and obese if their BMI was >30. The criteria for participation included the following: no dietary restrictions that could interfere with the experiments, including food allergies or religious or ethnic practices that limit food choice; medical conditions that could alter nutritional status or intestinal absorption (eg, inflammatory bowel disease); at least a moderate liking (>3 on a 5-point Likert-type scale) of the food used in the study and willingness to consume the food; and no psychopathology or developmental disabilities that would limit participation. We have never observed that sex moderates habituation in adults or children; however, to limit potential variability due to sex, only women were studied. All procedures and measures were approved by the University at Buffalo Social and Behavioral Sciences Institutional Review Board.

Design and procedure

After completing the phone screening, the participants were randomly assigned into 1 of 2 conditions: once a week sessions for 5 wk (weekly groups) or everyday sessions for 5 consecutive days (daily group). Within the obese and nonobese groups, 8 participants were randomly assigned to the weekly or daily groups. Visits were scheduled between the hours of 1100 and 1400. The subjects were asked to abstain from eating or drinking (except water) for 3 h before the study and from eating the study food, macaroni and cheese (Wegmans, Rochester, NY), 24 h before their appointment. On arrival to the laboratory, the participants were asked to read and sign a consent form. The participants completed a demographics form, same day food recall, and questionnaires regarding food preferences, hunger, and dietary restraint. The participants were told that they would participate in a laboratory research project that examines food preferences in adults, that the food to be studied was macaroni and cheese, and that the results of this study could help to further our understanding of changes in food preferences in adults. The participants were then instructed on the habituation task that they used during each visit (described below). At the completion of the experiment, the participants were debriefed about habituation processes and changes in food intake over time.

An intercom and closed-circuit video system were available so the experimenter could observe and communicate with the participant in the experimental room. Each test session was videotaped to ensure adherence to the study protocol. The participants were compensated \$15 per session, or \$75 total in the form of a Target gift card.

Measurement

Demographic variables and medical history

Educational level and racial-ethnic background were assessed by using a standardized questionnaire, whereas current medical problems, including psychiatric diagnoses and eating disorders, were assessed during an eligibility phone screen.

Weight, height, and BMI

Weight was assessed by using a Tanita BWB-800P digital scale (Arlington Heights, IL). Height was assessed with a Digit-Kit digital stadiometer (North Bend, WA). On the basis of the height and weight data, BMI was calculated. Participants were considered nonobese if their BMI was <30 and were considered obese if their BMI was ≥ 30 (14).

Subjective ratings of food liking and hunger

Participants provided subjective ratings of their hunger/fullness and liking of study foods before each session. Hunger was assessed by using a 5-point Likert-type scale anchored by "Extremely hungry" and "Extremely full." Liking of study foods was also determined by using a 5-point Likert-type scale anchored by "Do not like" and "Like very much."

Dietary restraint

Dietary restraint was measured with the Three-Factor Eating Questionnaire (TFEQ) (15). The TFEQ is a 40 item self-report questionnaire with true/false and multiple-choice questions. This measure assesses I) dietary restraint, 2) dietary disinhibition, and 3) hunger. Scoring >10 on the restraint scale will be considered to show dietary restraint.

Same-day food recalls

To ensure compliance with the protocol (not eating or drinking anything other than water for 3 h before testing), the participants recalled their dietary intake for that day.

Habituation task

The habituation task was a human analog to a basic animal research task used in studies on habituation to food (16, 17). The task was implemented during a 28-min period. The experimental room had 2 stations: 1 station contained a computer on which participants performed the food habituation task, and the other station contained a daily newspaper and sudoku and crossword puzzles, which were freely available if the participants no longer wanted to work for food on the habituation task. Having an alternative activity assured that participants were not just working to obtain food due to boredom.

Before beginning the habituation task, the participants were instructed on the task and given a practice period. The task provided participants the opportunity to earn access to food by clicking a mouse button. The participant could earn a portion of food contingent on clicking the mouse based on a variableinterval 120-s schedule (VI 120-s). Schedules of reinforcement are programs that determine the relation between responding for food and food availability and are used in habituation experiments (16). Interval schedules provide the opportunity for the participant to earn a point for the first button press after the interval is completed. A VI 120-s schedule provides the opportunity for the participants to earn a point on the average of once every 120 s, with a variation of ± 42 s (35%). The VI 120-s schedule was generated by using a computer program that determines reinforcement schedules. A random number generator was used to select the intervals within the schedule specifications. The VI 120-s was chosen based on pilot research comparing different VI schedules.

For every point earned, the participants received a 125-kcal portion (energy density = 1.89 kcal/g) of macaroni and cheese that was prepared according to the recipe on the package, which they could eat whenever they chose after it was earned. A batch (1750 kcal) of macaroni and cheese was prepared before the beginning of the experiment, and each 125-kcal portion of macaroni was provided in a small bowl and heated for 10 s before being presented to the participants. Responding during the 28min task was divided into fourteen 2-min time blocks. Trials to habituation was defined as the last 2-min trial in which responding for food was recorded before the first 2-min trial in which zero responding for food was recorded.

The experimental environment was set up for eating and olfactory experiments with an air-delivery system that continually cycles fresh air into each room. The rooms are in negative pressure, so that the exhaust has greater cubic feet per minute than the supply, and the air in the laboratory rooms is circulated ≈ 10 times/h. In addition, the experimental room (1385 cubic feet) includes a high-efficiency particle arresting (HEPA) filter that circulates air from 4.2 to 4.9 m³/min and contains a carbon potassium permanganate and zeolite filter to remove airborne odorants. The combination of removal of food stimuli after each trial, the air control system, and the HEPA filter reduce the possibility of smells lingering beyond an individual stimulus presentation.

TABLE 1

Baseline demographic characteristics of the participants¹

	Daily $(n = 8)$	Weekly $(n = 8)$	Daily $(n = 8)$	Weekly $(n = 8)$	Р
Age (y)	33.1 ± 12.2^2	32.8 ± 10.7	38.6 ± 7.6	38.7 ± 9.8	0.50
Weight (kg)	66.3 ± 7.0	66.8 ± 9.2	102.2 ± 16.2	103.2 ± 23.8	< 0.0001
BMI (kg/m^2)	24.6 ± 2.9	25.3 ± 3.5	37.7 ± 5.9	38.5 ± 7.9	< 0.0001
TFEQ score					
Disinhibition	5.1 ± 2.0	6.9 ± 2.5	10.3 ± 2.9	7.1 ± 3.6	0.01
Hunger	4.5 ± 2.6	6.9 ± 3.2	7.3 ± 3.1	4.8 ± 2.0	0.12
Restraint	12.0 ± 5.2	12.0 ± 4.3	9.6 ± 3.8	8.1 ± 4.9	0.27
Session 1 score					
Pre-liking	3.9 ± 0.6	4.0 ± 0.8	4.4 ± 0.7	4.0 ± 0.9	0.60
Pre-hunger	3.0 ± 0.8	2.0 ± 0.8	2.4 ± 0.6	2.6 ± 0.9	0.08
Trials to habituation	4.9 ± 4.1	3.9 ± 1.8	3.9 ± 1.1	4.0 ± 1.7	0.82
Energy intake (kcal)	476.2 ± 463.6	389.8 ± 108.7	427.0 ± 185.9	330.8 ± 161.3	0.74
Education (<i>n</i>)					
HS education	1	0	1	0	0.27
Some college or vocational training	5	5	6	8	
Completed 4 y of college or graduate school	2	3	1	0	
Income (<i>n</i>)					
<us\$50,000 td="" y<=""><td>5</td><td>4</td><td>4</td><td>2</td><td>0.42</td></us\$50,000>	5	4	4	2	0.42
≥US\$50,000/y	3	4	4	6	

Nonobese (n = 16)

¹ TFEQ, Three-Factor Eating Questionnaire; HS, high school. Differences between groups were tested by one-factor ANOVA for continuous variables and chi-square for categorical variables.

1

1

0

² Mean \pm SD (all such values).

Not of a minority race-ethnicity (*n*)

Data analysis

Differences between groups were tested by using one-factor analysis of variance for continuous variables or a chi-square test for categorical data. Changes in energy intake, trials to habituation, responding over the first 2 min of each session, and pre-session liking and hunger were analyzed by using mixed regression models (MRMs) (18) with random intercept and sessions. MRM models are ideally suited to habituation experiments because they compare slopes of the rate of change over sessions, incorporating individual-level heterogeneity (18). The relation between habituation and energy intake was assessed by using mixed regression with a random intercept that allows for testing relations across repeated measures (measures of responding or energy intake over days). MRMs also tested whether there were significant differences in the relation between the trials to habituation and energy intake by group and weight status. Characteristics of participants were correlated with energy intake and trials to habituation over days to identify variables that were considered as covariates in the MRM (19, 20). Analyses were performed by using the SAS (SAS Institute Inc, Cary, NC) and SYSTAT (Systat Software Inc, Chicago, IL) statistical programs.

RESULTS

The average participant had a mean (\pm SD) age of 35.8 \pm 10.1 y and BMI of 31.5 \pm 8.5. All but one participant had completed high school, whereas 6 (18.8%) had completed college. Fifteen of the participants had a family income <US\$50,000/y and 5 (15.6%) were minority. No differences were observed in any of these variables by group. Characteristics of participants by group are shown in Table 1. The only characteristic that was

Obese (n = 16)

0.21

3

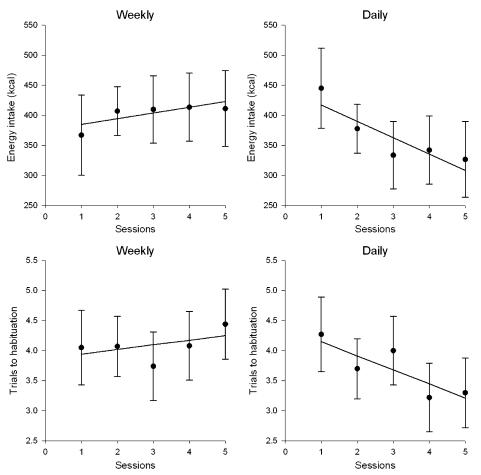


FIGURE 1. Mean (\pm SEM) energy intakes and trials to habituation for subjects assigned to once-weekly exposure of the same food for 5 wk (left graphs; n = 16) or daily exposure of the same food for 1 wk (right graphs; n = 16). Mixed-regression models showed a significant interaction by group over sessions for both energy (in kcal) consumed (P = 0.007) and trials to habituation (P = 0.049). The adjusted and the regression lines estimated from the mixed-regression models are shown. The regression estimates for energy intake were as follows: $\beta = -26.70$ (P = 0.014) and 8.97 (P = 0.36) for the daily and weekly groups, respectively, whereas the respective regression estimates for the trials to habituation were $\beta = -0.21$ (P = 0.067) and 0.05 (P = 0.70).

significantly different across groups was dietary disinhibition, which was included as a covariate in the MRM. The only individual difference variable that was reliably related to the trials to habituation or energy intake across sessions was income, and this variable was also included as a covariate in the MRM.

The MRM for energy intake showed that the interaction of group × session was significant ($F_{[1,96]} = 7.42$, P = 0.0077), because participants in the daily group had a reduction in energy intake over sessions ($\beta = -26.70$, P = 0.014), whereas those in the weekly group showed an increase ($\beta = 8.97$, P = 0.36) in energy intake over sessions (**Figure 1**). There was no weight status × session interaction (P = 0.35) or group × weight status × session interaction (P = 0.58).

The MRM for trials to habituation also showed a significant group × session interaction ($F_{[1,96]} = 3.97$, P = 0.049), but no weight status × session interaction (P = 0.17) or group × weight status × session interaction (P = 0.95). Participants in the daily group showed a reduction in trials to habituation over time ($\beta = -0.21$, P = 0.067), whereas those in the weekly group showed a small increase ($\beta = 0.05$, P = 0.70) in trials to habituation over sessions (Figure 1). No significant effects for group × weight status (P = 0.06), or a group × weight status

interaction (P = 0.79) were observed for initial responding during each session.

The MRM showed a significant effect of group on pre-session liking over sessions ($F_{[1,96]} = 4.14$, P = 0.045), because liking decreased over sessions for those in the daily group from 4.12 before session 1 to 3.75 before session 5 ($\beta = -0.14$, P = 0.025), whereas those in the weekly group showed a small increase in liking from session 1 to session 5 (4.0–4.06; $\beta = 0.006$, P =0.87). No effects of weight status (P = 0.35), or the interaction between group × weight status (P = 0.98) were observed on liking. Hunger ratings before each session showed no significant changes over session for group (P = 0.38), weight status (P =0.68), or the group × weight status interaction (P = 0.65).

The MRM showed that the number of trials to habituation predicted energy intake ($F_{[1,124]} = 57.79$, P < 0.0001), with greater number of trials to habituation associated with greater energy intake ($\beta = 49.39$, P < 0.0001). Group interacted with weight status to moderate the association between trials to habituation and energy intake ($F_{[1,124]} = 49.39$, P < 0.0001). The strongest relation was observed for nonobese subjects in the daily group ($\beta = 95.2$, P < 0.0001) in comparison with nonobese subjects in the weekly group ($\beta = 35.2$, P = 0.019) or obese

subjects in the daily ($\beta = 25.4$, P = 0.04) or weekly ($\beta = 46.9$, P < 0.0001) groups.

DISCUSSION

Whereas it is known that monotony, or repeatedly presenting the same food over days, will reduce food acceptability ratings and consumption (21–23), the results of this study provide the first evidence in humans that habituation may provide a theoretical explanation for why repeatedly consuming the same food will lead to reduced consumption. Long-term habituation, in terms of a faster rate of habituation and reduced energy intake, was observed for the daily group but not for the weekly group. Repeated presentations once a day compared with once a week provide a reference point for the interval between food presentations that could lead to long-term habituation. It is important to know how many days or meals can occur between food presentations and still observe long-term habituation. Parametric research is needed to determine whether there is a linear relation between the number of days and strength of long-term habituation or whether the effect is based on a threshold of how many days or meals can pass before long-term habituation would not be observed. One other important variable that may influence longterm habituation is the generalization across characteristics of the food stimulus (12). If foods are perceived as different, then the rate of long-term habituation over days or weeks will be reduced in comparison with foods that are perceived as the same. There is a question as to what constitutes the "same" food that would lead to long-term habituation (3). Research using the sensory-specific satiety paradigm has shown that small changes in food are enough to influence liking and consumption (24, 25), but these studies have involved foods presented within one meal or eating bout and not differences between foods over repeated meals over days. Will someone show long-term habituation to consecutive meals of cheese pizza, pepperoni pizza, and mushroom pizza? Likewise, would people show long-term habituation if they consumed macaroni and cheese and risotto with the same cheese sauce, because of the representations of the cheese sauce? It is likely that the level of habituation is based on generalization of characteristics of food across meals, but research is needed to identify what similarities across foods are adequate to produce long-term habituation.

It is of interest that obese subjects and nonobese subjects showed similar long-term habituation to daily presentations of the same food. Thus, whereas there are differences in habituation between obese and nonobese persons within the same meal (5, 7, 8), these differences did not extend to long-term habituation. These results suggest that repeated presentations of the same entrée over days would be equally effective for obese and nonobese women.

Long-term habituation is a memory phenomenon, and activation of memory traces from previous meals can enhance long-term habituation (1). Our model of habituation is based on extending Wagner's connectionist approach to memory, the Sometimes Opponent Processes (SOP) theory (26, 27) to habituation (1). The model hypothesizes that when a food stimulus is presented, a memory node for that stimulus is activated to its highest level, called the A1 state. The memory trace then decays over time to a lower level of activity, called the A2 state. Eventually, the memory trace further decays and becomes inactive. The model can be applied to habituation by hypothesizing that at the onset of the first food presentation, the memory node is activated to the A1 state, which quickly decays to A2. If the food stimulus is next presented when the memory node is still in the A2 state, reduced responding to the food stimulus will occur. However, if enough time has elapsed so that the memory trace becomes inactive, then the memory node can be reactivated to the A1 state again, which would lead to maximal responding for that food.

Long-term habituation involves associative conditioning (learning). During a later session, re-experiencing the context and sensory aspects that have been associated with food during previous sessions can activate the memory of food. However, when activated by such conditioned stimuli, the food node goes only to the A2 state. Thus, after a stimulus is experienced in a specific context, the conditioned contextual cues activate the habituating stimulus to the A2 state, which prevents full activation to the A1 state, providing a theoretical basis for long-term habituation. The fact that weekly macaroni and cheese (as opposed to daily macaroni and cheese) did not cause long-term habituation is consistent with the possibility that there is some forgetting of associative learning over the span of a week.

The major strength of the study is the application of habituation theory to understand how reducing the variety of foods over repeated meals can result in reduced consumption. However, the current study had several limitations. The study only involved premenopausal women, and, whereas we have not observed sex differences in any previous habituation studies, it is possible that memory differences for food exist between men and women, which could result in differences in long-term habituation. To our knowledge, there is no reason to hypothesize that menopause would influence habituation, and research on testing habituation of the acoustic startle responses across broad age ranges does not suggest that age predicts the rate of habituation (28). However, Rolls et al (25) showed that elderly participants have less sensoryspecific satiety than do younger participants, so it makes sense to assess habituation across wide age ranges in future research. The BMI values for the nonobese group were ≈ 25 , which includes both nonoverweight and overweight but not obese women. It is possible that collapsing across all women with a BMI < 30may obscure potential differences between overweight and nonoverweight women. The design, although it controlled for the number of stimulus presentations between groups, varied intervals between stimulus presentations. This resulted in the weekly group being studied over a longer time period than was the daily group, which may confound interpretation of the role of longterm habituation compared with the length of the time of the study. One way to equate the number of stimulus presentations and the total time being studied would be to have the daily group come to the laboratory for 20 consecutive weekdays and be served a variety of foods. During the last week, the subjects would be repeatedly served macaroni and cheese, whereas the weekly group would be served macaroni and cheese each Friday, while being served a variety of different foods on the other days.

Long-term habituation has many implications. Increasing food variety is a reliable way to increase energy intake within a meal (29), and increased variety in the diet is associated with greater body weight and poor choice of foods (29, 30). Reducing variety may be an important component of interventions for obesity (31–34). Habituation may provide a mechanism for the effects of variety on energy intake, such that within-session habituation

during a meal can lead to reduced intakes with reduced variety of foods (35). The long-term habituation reported here may be a mechanism for the effects of variety across (as opposed to within) meals. Thus, promoting long-term habituation by repeatedly serving the same food over days would lead to reduced energy intake over time. Such an intervention may be much simpler than the complex self-regulation approaches that are the basis for much of the current obesity therapy, which often meet with limited long-term success (36).

One important implication of these results is the potential for using memory and recall of eating as a way to reduce energy intake. In fact, recent research has shown that reminding someone of recent eating can decrease energy intake in a meal (37–39). Because increasing the rate of habituation is associated with reduced energy intake, the use of memory cues to recall recent eating may represent a unique complement to existing obesity treatments that bypass the need for extensive self-regulation to reduce eating. Habituation also may provide a unique perspective on how memory processes influence long-term energy intake.

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The authors' responsibilities were as follows—LHE: design of the experiment; KAC and MDC: supervision and collection of data; LHE, KAC, and RAP: statistical analysis and interpretation of data; LHE: drafting of the initial manuscript; and LHE, KAC, MDC, RAP, and MEB: critical revision of the manuscript. The study sponsors had no role in the writing of the report, in the decision to submit the manuscript for publication, or in the study design or collection, analysis, or interpretation of the data. LHE is a consultant for NuVal. None of the other authors declared a conflict of interest.

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