High Fat and Sugar Consumption During Ad Libitum Intake Predicts Weight Gain

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Objective: To determine how macronutrients accompanying foods with high energy density (EnDen) affect energy intake and weight gain.

Methods: A total of 214 subjects (130 males, BMI: 32 ± 7 kg/m²) ate ad libitum for 3 days. Food intake was expressed as the mean daily intake (in kilocalories) and the percentage of weight-maintaining energy needs (%WMEN). EnDen was expressed as the ratio of intake (in kilocalories) to food weight (in grams). Food choices were expressed as absolute and percent intake (kilocalories), categorized as high in fat (HF; ≥ 45% kcal) or low in fat (LF; < 20% kcal), and further categorized as high in complex carbohydrates (≥ 30% kcal), high in simple sugars (HSS; ≥ 30% kcal), or high in protein (HP; ≥ 13% kcal). Follow-up weights were available from 99 subjects (65 males, range: 6 months to 11 years).

Results: EnDen was associated with BMI (r = 0.28, P < 0.0001), percent body fat (r = 0.18, P = 0.007), and percent intake from HF/HP (r = 0.34, P < 0.0001), HF/HSS (r = 0.31, P < 0.0001), LF/HP (r = −0.37, P < 0.0001) and LF/HSS (r = −0.68, P < 0.0001). The %WMEN was associated with EnDen (r = 0.16, P = 0.01), HF/HSS (r = 0.33, P < 0.0001), and LF/HP intake (r = −0.25, P = 0.0002). In a multivariate model, only HF/HSS intake remained a significant predictor of %WMEN (β = 1.4% per 1% change, P < 0.0001). The percent intake from HF/HSS (r = 0.23, P = 0.02), not EnDen (P = 0.54), was associated with weight gain, even after adjusting for follow-up time (in years) and covariates.

Conclusions: Relatively greater consumption of HF/HSS foods independently predicted overeating and weight gain. Nutrient compositions of high-EnDen foods may be important for weight management.

Introduction

The wide availability of palatable and low-cost energy-dense foods may be a catalyst for increased energy intake and the increasing prevalence of obesity (1). Recent studies have shown that manipulating energy density (EnDen), a measure of calories per gram of food (2), is associated with increased energy intake (3) and subsequent weight gain (4). Thus, EnDen has been implicated as an important factor in weight management (5,6).

EnDen values of macronutrients range from 0 kcal/g (water) to 9 kcal/g (fat). Water content within foods accounts for some of the variability in EnDen values, although it provides no energy. Because of its high energy content, fat influences EnDen more than carbohydrates and protein (7,8). Thus, it has been proposed that targeting fat content would lead to lower energy intake (9). One study used three different methods to lower EnDen: (1) decreased fat content, (2) increased fruit/vegetable intake, or (3) increased water content. All led to reductions in intake, but intake was reduced the most by decreasing fat content (mean reduction of 396 kcal/d) compared to increased fruit/vegetable or water intake (10). However, other studies have reported that diets high in EnDen were associated with increased energy intake regardless of the dietary macronutrient composition (4). That is, all high-EnDen foods, whether due to high carbohydrate or high fat content, had the same effect on energy intake. Furthermore, when macronutrient composition of diets was kept constant (11), higher-EnDen diets led to overconsumption of food. Thus, whether manipulation of...
EnDen through macronutrient composition is sufficient to alter energy intake and subsequent weight is unclear.

Although there is evidence linking EnDen with longer-term energy intake and subsequent weight gain, the majority of studies have examined short-term changes in energy intake (at the current or subsequent meal) with prior or concurrent manipulation of EnDen (12,13). Furthermore, in the longer-term studies, the ingestion of high-fat foods, which tend to be high in EnDen, has often been underreported (14). The aim of the current study was to use an objectively acquired measure of EnDen via an ad libitum automated vending machine paradigm to investigate associations of EnDen and nutrient composition with measures of adiposity. Furthermore, we hypothesized that higher EnDen would be associated with increased weight gain and that this association would be driven by high-EnDen food groups.

Methods

Two hundred fourteen individuals aged 18 to 65 (BMI range: 18-52 kg/m²) without diabetes were recruited from the greater Phoenix, Arizona, area to participate in a larger study assessing eating behaviors and food preferences as risk factors for obesity (ClinicalTrials.gov identifier NCT00342732). Participants by race and/or ethnicity were represented as follows: 127 Native American (Southwestern heritage), 41 white, 12 Hispanic, 10 black, and 14 multiethnic participants. Prior to participation, all subjects were informed of the nature, purpose, and risks of the study and written informed consent was obtained. The protocol was approved by the institutional review board of the NIDDK. All subjects were found to be healthy based on medical history, physical examinations, and laboratory tests. Any evidence of acute or chronic disease was basis for exclusion from participation in the study.

Upon admission to the Obesity and Diabetes Clinical Research Unit (Phoenix, Arizona), subjects were fed a weight-maintaining diet with a macronutrient distribution of 50% carbohydrate, 30% fat, and 20% protein for 3 to 5 days prior to ad libitum food consumption. Weight maintenance calories were calculated for each subject based on weight, gender, and BMI, as previously described (15). Body composition was determined by dual-energy x-ray absorptiometry (Prodigy; General Electric Healthcare, Chicago, Illinois). Subjects underwent an oral glucose tolerance test to confirm nondiabetes status on day 3 of admission.

Follow-up visits

Data from return visits or from re-enrollment into other studies on our research unit with assessment of body composition measures were used to assess changes in adiposity over time. Subjects’ last available return or re-enrollment visit was used as the follow-up.

Vending machine paradigm

Ad libitum food intake was measured by using automated vending machines, as previously described (16-18). Previous studies on our unit have shown high reproducibility in intraperson energy intake patterns during repeated visits (16). This method provides a highly accurate assessment of energy intake for persons confined in a research unit. After admission to the metabolic ward, subjects completed an 80-item Food Preferences Questionnaire containing typical breakfast, lunch, dinner, and snack items, as described by Geiselman et al (19). Subjects were asked to rate their preference for a wide variety of foods on a nine-point Likert scale. Forty different foods given an intermediate rating were used to stock the vending machines for ad libitum food intake. These 40 foods were available each of the 3 days along with condiments (Supporting Information Figure S1). Subjects were given free access to the machines for 23.5 hours with 30 minutes per day needed to restock the machine. The machines were computer operated and required a unique code, which was given to each subject for access. The machines recorded the time of day each food was accessed. These data were then imported into Food Processor SQL Edition (version 10.0.0; ESHA Research, Salem, Oregon) or the CBORD Professional Diet Analyzer Program (version 4.1.11, CBORD Inc., Ithaca, New York), which provided the calories and macronutrient content of the foods (Supporting Information Figure S1). EnDen (in kilocalories per gram) was calculated as the average calories consumed over the 3 days of ad libitum feeding divided by the weight (in grams) of food eaten per day. Weights of foods were taken from Food Processor and CBORD output. In order to correct for actual food intake, the metabolic kitchen staff weighed food leftovers returned by the individuals. Beverage consumption was included in the EnDen calculations.

Macronutrient categories

Each food on the vending questionnaire was categorized into one of six groups (Supporting Information Figure S2), based on the macronutrient content as a percentage of the total energy. Foods were categorized as high in fat (HF; ≥45% kcal) or low in fat (LF; <20% kcal) and then further categorized as being high in complex carbohydrates (HCC; ≥30% kcal), high in simple sugars (HSS; ≥30% kcal), or high in protein (HP; ≥13% kcal). These categorizations produced six different groups of food: HF/HCC, HF/HP, HF/HSS, LF/HCC, LF/HP, and LF/HSS (19). The raw energy intake (in kilocalories) from each food group was further converted to the percentage of total energy intake.

Statistical analysis

Statistical analysis was done using SAS (version 9.3) and SAS Enterprise Guide (version 5.1) (SAS Institute Inc.). Alpha was set at 0.05 for all analyses. Normally distributed data are presented as mean ± SD, whereas nonparametric data are presented as median (interquartile range). Gender and racial differences for anthropometric data were assessed by using Student t tests, and differences between categorical variables were analyzed by using χ² tests. There were no differences among individuals who were white, Hispanic, black, or from another racial background (n = 87); thus, we combined these individuals to compare Native American participants (n = 127) with individuals of all other racial groups. EnDen is reported as the average calories eaten over 3 vending days divided by the average food weight eaten over the same 3 days. Differences in absolute average daily energy intake from each of the six groups and the percent daily energy intake from each food group were assessed by using analysis of variance (ANOVA). Analysis of covariance (ANCOVA) was used to assess the absolute average daily energy intake from each of the six groups adjusted for total average daily energy intake. Post hoc pairwise comparisons with Bonferroni adjustments were used for significant outcomes. Differences in
baseline values between subjects with and without follow-up data were analyzed by using the Student t test. Pearson correlation coefficients were used to assess relationships between normally distributed variables, and Spearman correlation coefficients were used for skewed data. The change in weight was assessed by calculating the difference between baseline and follow-up weight. Thus, a positive change in weight is representative of weight gain. Multivariate linear regression analyses were adjusted for age, sex, and race. Similar associations were observed in the cross-sectional analyses, where EnDen was positively associated with %WMEN, whereas the percent intake from LF/HP was negatively associated with %WMEN. The percent intake from HF/HP was positively associated with %WMEN, whereas the percent intake from HF/HSS was negatively associated with %WMEN.

### Results

Subject characteristics are shown in Table 1. Men had lower percent body fat, fat mass (P < 0.0001), and BMI and had higher fat-free mass. Men had higher energy intakes and overate a larger percent of their weight-maintaining energy needs. Native American participants had higher percent body fat (P = 0.001) and fat mass (P = 0.01), tended to be younger (P = 0.05), and had higher energy intakes (P = 0.02). The average percentage of weight-maintaining energy needs (%WMEN) was 148%, suggesting that the majority of individuals overate on the vending machine paradigm, which is consistent with findings from the literature (16,19). The %WMEN was calculated by dividing the total average calories by the weight-maintaining energy needs and expressed as a percentage (Table 1, Figure 1).

In the cross-sectional analyses, EnDen was positively associated with weight (r = 0.23, P = 0.0008), BMI (r = 0.28, P < 0.0001), percent body fat (r = 0.18, P = 0.007), fat mass (r = 0.23, P = 0.007), fat mass index (r = 0.24, P = 0.0005), fat-free mass (r = 0.15, P = 0.03), and fat-free mass index (r = 0.26, P = 0.0002). These relationships remained unchanged after adjustment for age, sex, and race. EnDen was positively correlated with the percent daily energy intake from HF/HP (r = 0.34, P < 0.0001; Figure 1A) and HF/HSS (r = 0.31, P < 0.0001; Figure 1B) and negatively correlated with the percent daily energy intake from LF/HP (r = -0.37, P < 0.0001; Figure 1C) and LF/HSS (r = -0.68, P < 0.0001; Figure 1D). There were no associations with HF/HCC (r = 0.10, P = 0.14) and LF/HCC (r = -0.02, P = 0.75). These relationships were still significant after adjustments for age, sex, and race. Similar associations were found for absolute daily energy intake from the six food groups as well as with absolute food intake from each group adjusted for total daily energy intake.

### Table 1 Participant demographics

| Variable                      | All          | Male         | Female        |
|-------------------------------|--------------|--------------|---------------|
| N                             | 214          | 130          | 84            |
| Race                          | 127 NA; 87 O | 71 NA; 59 O  | 56 NA; 28 O   |
| Age                           | 35 ± 9       | 35 ± 9       | 34 ± 9        |
| Weight (kg)                   | 90 ± 21      | 92 ± 20      | 87 ± 22       |
| BMI (kg/m²)                   | 32 ± 7       | 30 ± 6       | 34 ± 8        |
| Percent body fat (%)          | 31 ± 9       | 26 ± 7       | 38 ± 6        |
| FM (kg)                       | 29 ± 12      | 25 ± 11      | 34 ± 12       |
| FMI (kg/m²)                   | 10 ± 5       | 8 ± 3        | 13 ± 5        |
| FFM (kg)                      | 61 ± 13      | 67 ± 11      | 53 ± 10       |
| FFMI (kg/m²)                  | 21 ± 4       | 22 ± 3       | 20 ± 4        |
| Fasting glucose concentration (mg/dL) | 91 ± 8   | 91 ± 8       | 92 ± 8        |
| 2-h glucose concentration (mg/dL) | 125 ± 29  | 121 ± 27     | 129 ± 29      |
| Energy intake (kcal)          | 4,108 ± 1,369| 4,540 ± 1,265| 3,440 ± 1,258 |
| Food weight (g)               | 2,967 ± 984  | 3,281 ± 901  | 2,480 ± 909   |
| %WMEN                         | 148 ± 46     | 158 ± 41     | 132 ± 48      |
| %HF/HCC                       | 12 ± 8       | 13 ± 8       | 13 ± 7        |
| %HF/HP                        | 28 ± 11      | 28 ± 10      | 27 ± 11       |
| %HF/HSS                       | 17 ± 9       | 18 ± 9       | 17 ± 10       |
| %LF/HCC                       | 8 ± 6        | 8 ± 5        | 9 ± 7         |
| %LF/HP                        | 11 ± 6       | 11 ± 6       | 11 ± 7        |
| %LF/HSS                       | 18 ± 7       | 18 ± 7       | 18 ± 8        |

*Sex differences, P < 0.05.*

EnDen, energy density; FFM, fat-free mass; FFMI, fat-free mass index; FM, fat mass; FMI, fat mass index; HF, high in fat; HCC, high in complex carbohydrates; HP, high in protein; HSS, high in simple sugars; LF, low in fat; NA, Native American; O, other; %WMEN, percentage of weight-maintaining energy needs.
intake from HF/HSS, and percent intake from LF/HP, only percent intake from HF/HSS ($\beta = 1.4\%$ per 1\% change in HF/HSS, 95\% CI: 0.76-2.1, $P < 0.0001$) remained a significant predictor of %WMEN.

ANOVA revealed significant differences in absolute daily energy (in kilocalories per day) consumed from the six food groups ($P < 0.0001$; Figure 2). Post hoc pairwise comparisons with Bonferroni adjustments reported that the highest daily energy intake was from the HF/HP food group (mean ($M = 1,141$, 95\% CI: 1,097-1,185)). Individuals consumed relatively similar amounts of energy from the HF/HSS ($M = 770$, 95\% CI: 726-815) and LF/HSS groups ($M = 715$, 95\% CI: 671-760), as well as similar amounts from the HF/HCC ($M = 538$, 95\% CI: 48-591) and LF/HP food groups ($M = 439$, 95\% CI: 394-482). The lowest daily energy intake was from the LF/HCC food group ($M = 318$, 95\% CI: 274-362). Results were identical for the ANCOVA model using absolute energy intake from each food group adjusted for total average daily energy intake.

There were significant differences in percent daily energy consumed from the six food groups ($P < 0.0001$). Post hoc pairwise comparisons with Bonferroni adjustments reported the highest percent daily energy intake was from the HF/HP food group ($M = 27.5\%$, 95\% CI: 26.5-28.6). Individuals ate relatively similar amounts of energy from the HF/HSS ($M = 17.4\%$, 95\% CI: 16.3-18.5) and LF/HSS ($M = 17.8\%$, 95\% CI: 16.8-18.9) food groups as well as similar amounts from the HF/HCC ($M = 12.7\%$, 95\% CI: 11.7-13.8) and LF/HP food groups ($M = 11.3\%$, 95\% CI: 0.3-12.4). The lowest percent daily energy intake was from the LF/HCC food group ($M = 8.4\%$, 95\% CI: 7.4-9.5; Figure 2).

Ninety-nine subjects had follow-up data available. In a comparative analysis between subjects with follow-up data and subjects without follow-up data, there were no significant differences on continuous or categorical variables (Table 2). EnDen itself ($r = 0.06$, $P = 0.54$), %WMEN ($r = 0.07$, $P = 0.48$), and the absolute daily energy intake from all six food groups ($P > 0.05$) were not associated with
changes in weight. However, absolute daily energy intake from the HF/HSS food group adjusted for total daily energy intake approached significance with weight gain ($r = 0.19$, $P = 0.06$). Similarly, energy intake from those who ate a greater percentage of their calories from the HF/HSS food group led to more weight gain ($r = 0.23$, $P = 0.02$; Figure 3). In a multivariate linear model with weight change as the dependent variable, adjusted for age, sex, race, and follow-up time, the percentage of calories from the HF/HSS food group predicted weight gain ($\beta = 0.18$ kg per 1% change in HF/HSS, 95% CI: 0.03-0.33, $P = 0.03$, $\eta^2 = 0.06$). Results using the absolute calories from the HF/HSS food group adjusted for total daily calories instead of expressed as a percentage resulted in similar results ($\beta = 0.4$ kg per 100-kcal change in HF/HSS, 95% CI: 0.001-0.008, $P = 0.03$, $\eta^2 = 0.06$). Results did not change with adjustment for baseline weight (data not shown).

**Discussion**

In the current study, we have shown that intake from HF/HSS foods, rather than EnDen, independently predicts overeating and weight gain. Individuals who consumed foods with greater overall EnDen had higher adiposity, consumed a greater proportion of energy (in kilocalories) from HF/HP and HF/HSS food groups, and consumed a lower proportion of energy from LF/HP and LF/HSS food groups. Although EnDen and intake from HF/HSS food groups were both associated with overeating, in a model containing all significant

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**TABLE 2 Follow-up demographics**

|                  | Baseline | Follow-up |
|------------------|----------|-----------|
| N                | 214      | 99        |
| Sex              | 130 males; 84 females | 65 males; 34 females |
| Race             | 127 NA; 87 0 | 62 NA; 37 0 |
| Age              | 35 ± 9   | 35 ± 9    |
| Weight (kg)      | 90 ± 21  | 89 ± 21   |
| Weight change (kg) | 2.7 ± 7.3 | 858 (384, 1,613) |
| Follow-up time (d) | 32 ± 7   | 125 ± 29  |
| BMI (kg/m²)      | 31 ± 7   | 29 ± 9    |
| Percent body fat | 31 ± 9   | 27 ± 13   |
| FM (kg)          | 29 ± 12  | 10 ± 5    |
| FMI (kg/m²)      | 11 ± 4   | 62 ± 12   |
| FFM (kg)         | 61 ± 13  | 21 ± 4    |
| FFMI (kg/m²)     | 21 ± 4   | 92 ± 8    |
| Fasting glucose concentration (mg/dL) | 91 ± 8 | 123 ± 28 |
| 2-h glucose concentration (mg/dL) | 125 ± 29 | 123 ± 28 |
| EnDen (kcal/g)   | 1.4 ± 0.3 | 1.4 ± 0.3 |
| Energy intake (kcal) | 4,108 ± 1,369 | 4,243 ± 1,355 |
| Energy volume (g) | 2,967 ± 984 | 3,044 ± 925 |
| %WMEN            | 148 ± 46 | 154 ± 46  |
| %HF/HCC          | 12 ± 8   | 13 ± 8    |
| %HF/HP           | 28 ± 11  | 27 ± 9    |
| %HF/HSS          | 17 ± 9   | 17 ± 9    |
| %LF/HCC          | 8 ± 6    | 8 ± 6     |
| %LF/HP           | 11 ± 6   | 11 ± 6    |
| %LF/HSS          | 18 ± 7   | 18 ± 7    |

Apart from N, race, and age, all data presented as mean ± SD or median (25% IQR, 75% IQR). EnDen, energy density; FFM, fat-free mass; FFMI, fat-free mass index; FM, fat mass; FMI, fat mass index; HF, high in fat; HCC, high in complex carbohydrates; HP, high in protein; HSS, high in simple sugars; IQR, interquartile range; LF, low in fat; NA, Native American; O, other; %WMEN, percentage of weight-maintaining energy needs.
High Fat and Sugar Intake Predicts Weight Gain

Obesity conditions in a research unit, using Our results broaden the existing knowledge of EnDen to free-living EnDen itself, may be more important for weight management. that the nutrient composition of energy-dense foods, rather than foods, but not EnDen, predicted weight gain. These findings indicate that the nutrient composition of energy-dense foods, rather than EnDen itself, may be more important for weight management.

Our results broaden the existing knowledge of EnDen to free-living conditions in a research unit, using ad libitum conditions utilizing an objective measure of energy intake (19). This ad libitum vending machine system has been shown to be valid and highly reliable within individuals during repeated trials (16,19,20). In contrast, other research has examined intake using less reliable measures such as self-reported dietary intake or 24-hour food recalls, which lead to underreporting of energy intake and biased food reports (21-23). Furthermore, our utilization of six different nutrient mixtures allowed us to identify a potential mechanism by which energy-dense foods may drive caloric intake. The positive association between intake from HF/HP and HF/HSS and the inverse association between LF/HP and LF/HSS food groups and EnDen support fat content as an important factor determining EnDen (4). However, only intake from HF/HSS food groups predicted overeating (%WMEN) and weight gain. Thus, although we found associations between EnDen and adiposity measures, it was the specific nutrient mix of high-EnDen food (i.e., HF/HSS) that determined both overeating and weight gain, thus indicating a mechanism by which energy-dense foods may drive caloric intake.

The current study builds upon existing literature, which has suggested that the quality of the nutrient mix of fat and sugar intake adversely affects energy intake through reward mechanisms (10,24). Neuroimaging studies have found that both high-fat and high-sugar foods activate reward areas within the brain (25). Rodent models have found that rats fed high-fat and high-sugar diets are motivated to obtain sugar rewards even when satiated (26). Furthermore, studies examining energy intake have reported that sugar is associated with overeating because of its desirable sweet taste (27,28) as well as its association with impaired cognitive function (29). Specifically, high fat and sugar intake is related to poorer hippocampal function, which leads to less accurate memories of prior food intake as well as reduced hunger and satiety signals. Thus, our finding that HF/HSS food was associated with greater energy intake and predicted weight gain is consistent with findings from previous studies.

Some limitations of the current study should be acknowledged. First, the vending machine paradigm is an artificial environment with no limitations or barriers to food consumption, which leads to facilitation of overeating. However, the vending machine paradigm yields greater reliability of food intake compared to traditional self-report assessments (16,30). Secondly, our longitudinal analysis did not include everyone who was studied. However, there were no significant differences between those who had follow-up and those who did not. Thirdly, the average BMI at baseline and follow-up was in the obesity range (≥ 30), which may have led to biases in the associations between adiposity measures and EnDen.

Conclusion

In summary, we demonstrated that EnDen is associated with higher levels of adiposity, greater relative consumption of HF/HP and HF/HSS foods, and lower consumption of LF/HP and LF/HSS foods. EnDen and consumption of a relatively larger percentage of HF/HSS foods were associated with overeating. However, in adjusted models, only the percent intake from HF/HSS foods predicted overeating. In longitudinal analysis, greater consumption of HF/HSS foods specifically, rather than overall EnDen, %WMEN, or other high-EnDen food groups (i.e., HF/HP), predicted weight gain. Thus, within foods with greater EnDen, overall nutrient composition appears to be important in determining overeating and weight gain.

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