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ORIGINAL ARTICLE

A reciprocal interaction between food-motivated behavior and diet-induced obesity

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Objectives: One of the main causes of obesity is overconsumption of diets high in fat and sugar. We studied the metabolic changes and food-motivated behavior when rats were subjected to a choice diet with chow, lard and a 30% sucrose solution (high fat high sugar (HFHS)-choice diet). Because rats showed considerable variations in the feeding response to HFHS-choice diet and in food-motivated behavior, we investigated whether the motivation to obtain a sucrose reward correlated with the development of obesity when rats were subsequently subjected to HFHS-choice diet.
Method: We first studied feeding, locomotor activity and body temperature, fat weights and hormonal concentrations when male Wistar rats were subjected to HFHS-choice diet for 1 week. Second, we studied sucrose-motivated behavior, using a progressive ratio (PR) schedule of reinforcement in rats that were subjected to the HFHS-choice diet for at least 2 weeks, compared to control rats on a chow diet. Third, we measured motivation for sucrose under a PR schedule of reinforcement in rats that were subsequently subjected to HFHS-choice diet or a chow diet for 4 weeks. Fat weights were measured and correlated with the motivation to obtain sucrose pellets.
Results: One week on the HFHS-choice diet increased plasma concentrations of glucose and leptin, increased fat stores, but did not alter body temperature or locomotor activity. Moreover, consuming the HFHS-choice diet for several weeks increased the motivation to work for sucrose pellets. Furthermore, the motivation to obtain sucrose pellets correlated positively with abdominal fat stores in rats subsequently subjected to the HFHS-choice diet, whereas this correlation was not found in rats fed on a chow diet.
Conclusion: Our data suggest that the motivation to respond for palatable food correlates with obesity due to an obesogenic environment. Conversely, the HFHS-choice diet, which results in obesity, also increased the motivation to work for sucrose. Thus, being motivated to work for sucrose results in obesity, which, in turn, increases food-motivated behavior, resulting in a vicious circle of food motivation and obesity.
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Keywords: abdominal obesity; food-motivated behavior; progressive ratio schedule of reinforcement; choice diet; lard; sucrose

Introduction

Obesity is one of the fastest growing medical problems in modern society. It contributes to the development of chronic disorders such as cardiovascular disease, type 2 diabetes, hypertension and some cancers.[1] For healthy body weight regulation, it is important to balance energy intake and energy expenditure. Modern society, however, is characterized by a sedentary lifestyle and a calorie intake, which is not adjusted for that. Although regulatory mechanisms that balance intake and expenditure are known ,[2,3] it still remains unclear why this fails in obese individuals. It has been postulated that the ability to overconsume high-energy dense foods (in the face of plentiful food availability) was selected during evolution. Thus, under conditions when food availability is uncertain and scarce, consuming high-energy dense foods is an adequate adaptation (for a review see Ulijazek[4]). Furthermore, children and adults have preferences for foods associated with fat and sugar,[5,6] and the increased intake of saturated fat and sugar-based beverages has been linked to obesity.[7] It is, therefore, important to study further the regulation of consumption of dietary fat and sugar in solution and their role in the development of obesity.[8–13] Furthermore, these studies should include choices made daily by people living in

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wealthy societies in which different high-energy foods are readily available. Many animal models have been developed to study the involvement of high-fat/ high-energy diets in the development of obesity. The diets used in these studies, however, consist of pellets that contain all nutrients in one pellet.[14,15] We, therefore, started subjecting rats to a free choice of different palatable items, in addition to the normal balanced rat chow.[16,17]

When rats were subjected to lard and a 30% sucrose solution in addition to normal balanced chow and water, they increased (abdominal) fat stores and plasma leptin concentrations because of an increase in total calorie intake.[16,17] This overconsumption of palatable food is probably due to its hedonic properties. Indeed, even sated rats will work hard for fat and sugar.[18] The motivation to obtain a reward (e.g. sugar) can be tested using a progressive ratio (PR) schedule of reinforcement.[19,20] Under a PR schedule, the cost of a reward is progressively increased over successive trials to determine how hard the rat is willing to work for it. The so-called breakpoint, that is, the maximal amount of responses an animal makes for a single reward, is used as a measure of the incentive value of the reward.

In the seventies food-motivated behavior in relation to obesity has been studied extensively.[21–25] Several genetically obese animals show an increased motivation for food as measured under different reinforcement schedules. However, in nongenetic obese models, changes in food-motivated behavior were not observed.[22–26] It is, therefore, unclear whether there is an effect of obesity on food-motivated behavior, and to what extent such behavior affects the development of obesity.

The present experiments were designed to characterize the metabolic changes and food-motivated behavior when rats were subjected to a choice diet with chow, lard and a 30% sucrose solution. First, we studied feeding, locomotor activity and body temperature, fat weights and hormonal concentrations when rats were subjected to the choice diet for 1 week to demonstrate the development of obesity. Second, we studied food-motivated behavior using a PR schedule of reinforcement in rats that were subjected to the high fat high sugar (HFHS)-choice diet for at least 2 weeks, compared to control rats on a chow diet. Because rats showed considerable individual variation in their response to the HFHS-choice diet and the amount of active lever presses under the progressive ratio, we asked whether this motivation to obtain a reward is predictive of developing obesity when rats were subsequently subjected to the HFHS-choice diet.

Materials and methods

Animals

Male Wistar rats (Charles River, Sulzfeld, Germany) weighing 220–250 g at the beginning of the experiment were individually housed in macrolon cages with *ad libitum* access to rat chow (SDS special diet service, England) and water. Animals were kept in a temperature- and humidity-controlled room ($21 \pm 2^{\circ}C$) under a 12/12 h light/dark cycle (Experiment 1: lights on at 0700; Experiment 2: lights on at 1900). All experimental procedures were approved by the Committee for Animal Experimentation of the University Medical Center Utrecht, the Netherlands.

Experiment 1

Surgery. Twenty rats received transmitters (TA10TA-F40, Data Sciences International, St Paul, MN, USA) in the abdominal cavity under fentanyl/fluanisone (Hypnorm, Janssen Pharmaceutica, Beerse, Belgium; 0.1 ml/100 g i.m.) and midazolam anesthesia (Dormicum, Hoffman-La Roche, Mijdrecht, The Netherlands; 0.05 ml/100 g i.p.). After surgery, rats were treated with carprofen (5 mg/kg s.c.) and saline (min 3 ml s.c.) and were allowed to recover for a week.

Experimental design. Two weeks after surgery, 10 rats were switched to a high-fat, high-sucrose (HFHS)-diet: a dish of lard and a bottle of 30% sugar water were present in the cage in addition to their normal standard chow and water bottle. The other rats remained on *ad libitum* chow and water. Seven days after the start of the HFHS-choice diet, all rats were killed between 0900 and 1000 by decapitation within 10 s after they had been taken from their home cages.

Data analysis. Food intake (chow, lard and sugar water) and body weight were measured daily from 1 week after surgery until the end of the experiment. Body temperature and locomotor activity were measured during 5 days before and during the week when rats were subjected to the HFHS-choice diet (or the control condition). The transmitters sent digitized data via radio frequency signals to a nearby receiver. These data were automatically recorded every 10 minutes, and averaged per hour using DSI software (DSI, St Paul, MN, USA). Data from two HFHS-choice diet rats and two control rats were excluded from analysis because of incomplete measurements. From the trunk blood, blood glucose and plasma concentrations of insulin and leptin were determined. Individual mesenteric, epididymal, s.c.(inguinal) and perirenal white adipose tissues, thymus and adrenals were dissected, cleaned and weighed.

Plasma measurements. Blood glucose was measured in duplicate using a Medisense glucosesensor (Abbott, Amersfoort, The Netherlands). Plasma leptin and insulin were analyzed in duplicate using radioimmunoassay kits (Linco Research, St Charles, MI, USA).

Experiment 2

At least 7 days after the arrival of the rats from the supplier, food intake, water intake and body weight were measured for several days to determine baseline feeding behavior. Eight

rats were subjected to HFHS-choice diet as described above (day 0). This HFHS-choice diet was given for the rest of the experiment. Intake of chow, lard and the sucrose solution was measured daily. A second group ($n=8$) was kept on normal standard chow diet and served as a control group.

Apparatus. Operant conditioning experiments took place in two-lever operant conditioning chambers designed for rats ($30.5 \times 24.1 \times 21.0$ cm; Med-Associates, Georgia, VT, USA), which were placed in sound-attenuating chambers. Each chamber had a metal grid floor, two retractable levers with white light bulbs above it and a food-pellet dispenser that could deliver 45 mg sucrose pellets (Noyes Precision Pellets Formula F, Research Diets, New Bruncswick, NJ, USA) to the food tray. Each chamber was illuminated by a white light bulb. Data collection and processing was controlled by MED-PC software.

Training. Starting on day 14, rats learned to lever press for sucrose pellets during 7 sessions under a fixed ratio (FR)1 schedule, with 2 sessions/day. A single press on the active lever resulted in the delivery of one sucrose pellet, illumination of the light above the lever and retraction of the lever. Twenty seconds after the pellet was received, the lever was reinserted into the chamber. Sessions lasted 30 min or until the rats earned 60 pellets, whichever occurred first. Presses on the inactive lever were recorded, but had no programmed consequence. Seven FR1 schedule sessions were followed by the progressive ratio (PR) schedule, which started on day 21. Under a PR schedule, the cost of a reward is progressively increased over successive trials, to determine the effort the rat will emit for it. The response requirement increased according to the following equation[19,20,27]: response ratio $=(5e(0.2 \times \text{infusion number})) - 5$ through the following series: 1, 2, 4, 9, 12, 15, 20, 25, 32, 40, 50, 62, 77, 95, 118, 145, 178, 219, 268, 328, 402, 492, 603, 737. The session ended when the rat had failed to earn a reward within 60 min. Responding was considered stable when the number of food pellets earned per session did not differ more than 15% for three consecutive sessions. In most cases, responding stabilized within 5–8 sessions. Rats were tested for a total of 11 sessions, with 1 session/day. The experiment ended on day 33 (rats were not tested on days 19 and 20).

Experiment 3

At least 7 days after the arrival of the rats from the supplier, food intake, water intake and body weight were measured for several days to determine baseline feeding behavior. Next, rats were trained to lever-press for sucrose pellets as described in Experiment 2. After training under the FR schedule, all rats were switched to the PR schedule and tested for 10 sessions with one session/day. Two days after the last PR session, 18 rats were switched to HFHS-choice diet, whereas 12 rats remained on the chow diet. This experiment was performed in two runs. Four weeks after starting the HFHS-choice diet, rats were killed between 0900 and 1000 by decapitation within 10 s after they had been taken from their home cages and trunk blood was collected. Individual mesenteric, epididymal, s.c.(inguinal) and perirenal white adipose tissues, thymus and adrenals were dissected, cleaned and weighed.

Statistical analysis

Metabolic data were analyzed using Student's *t*-tests. The behavioral data and body temperature were analyzed with a repeated-measures analysis of variance (ANOVA) with diet as between subjects factor and sessions or time as within-subjects factor. When significant overall interactions were found, *post hoc* analyses were performed with a Student's *t*-test. Simple linear regression analyses were performed to determine the relationship between variables. All data were analyzed with SPSS software. A result was considered significant if $P<0.05$. All data are expressed as mean $\pm$ s.e.m.

Results

The HFHS-choice diet

When switched to the HFHS-choice diet, rats consumed a stable amount of chow, lard and 30% sucrose solution over the 7 days. Total calorie intake over 7 days was significantly higher in HFHS-choice diet rats, compared to the intake of control rats (Figure 1a $P<0.02$). HFHS-choice diet rats consumed 47.8% ($\pm$2.6) of their calories from chow, 37.4% ($\pm$4.3) from lard, and 14.8% ($\pm$2.1) from sucrose (Figure 1b). For calorie intake, the variance between rats within the HFHS-choice diet group was considerably larger compared to the chow-fed rats (Levens test: $P=0.02$), indicating variation in the response to the HFHS-choice diet. One week of HFHS-choice diet significantly increased all fat stores measured (Figure 1c, $P<0.05$), although there was no effect on weight gain (Figure 1d). Body temperature and locomotor activity were not affected by the HFHS-choice diet (Figure 2). Plasma leptin concentrations were significantly increased (Table 1). Abdominal (epidydimal, perirenal and mesenteric) fat stores correlated positively with plasma leptin concentrations in HFHS-choice diet rats ($R^2=0.85$; $P<0.001$), whereas they did not correlate with abdominal fat stores in control rats (data not shown). Blood glucose concentrations were increased in HFHS-choice diet rats compared to controls; however, plasma insulin concentrations were similar. Adrenal and thymus weight were similar in both groups of rats (Table 1).

Effects of the HFHS-choice diet on food-motivated behavior

During the 2 weeks before training, rats subjected to the HFHS-choice diet consumed more calories compared to the chow control group (Figure 3a, $P<0.01$). The HFHS-choice diet rats consumed 50.2% ($\pm$2.1) of their calories from chow, 37.4% ($\pm$2.9) of their calories from lard and 12.5% ($\pm$1.7) of

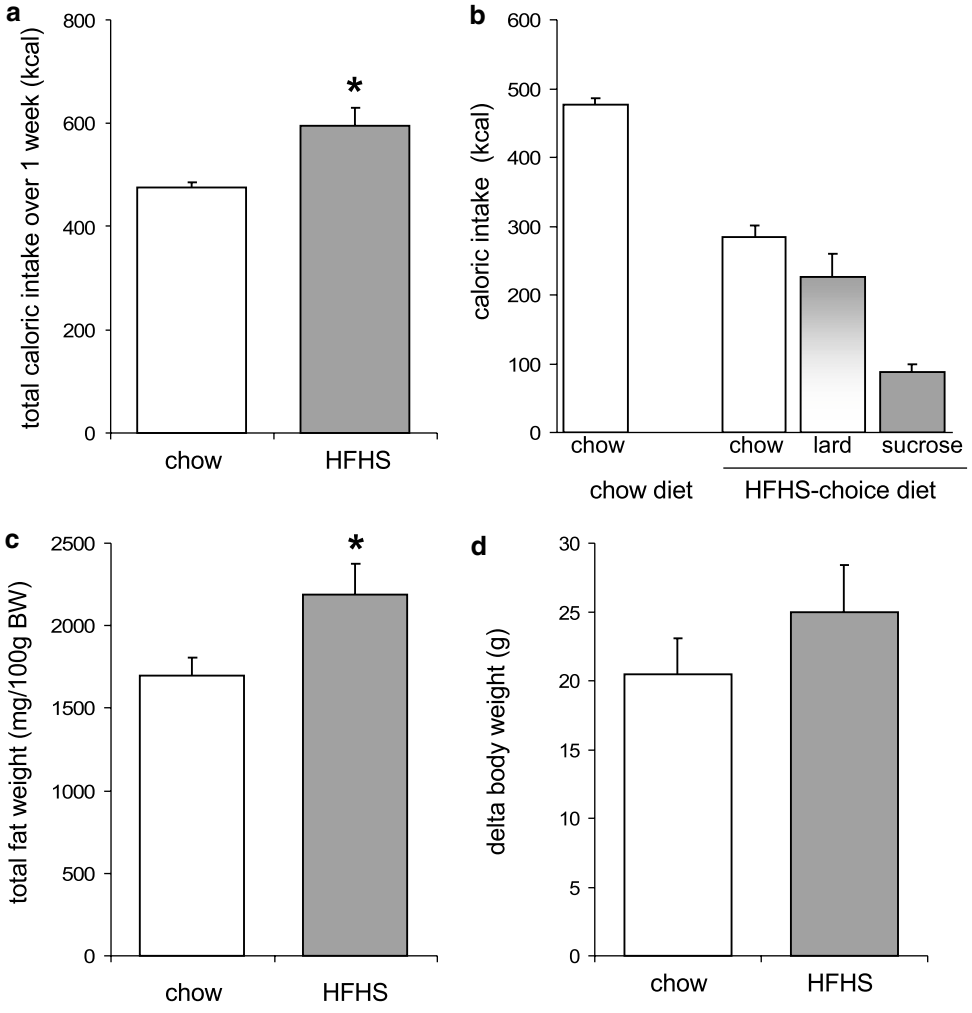

Figure 1 (**a**) Total calorie intake over 7 days was higher in HFHS-choice diet rats compared with chow-fed rats. (**b**) Calories from chow, lard and the 30% sucrose solution in rats on HFHS-choice diet over 7 days. (**c**) The HFHS-choice diet resulted in increased total fat stores. (**d**) No differences were found in delta body weight over 1 week on the HFHS-choice diet. White bars: chow-fed. Gray bars: HFHS-choice diet. *: $P<0.05$. Data are mean ± s.e.m. ($n=6$).

their calories from sucrose. Body weight, however, did not change within these 2 weeks (Figure 3b).

After 2 weeks, rats were tested under the FR schedule for seven sessions. Figure 4 shows the amounts of rewards obtained under the FR1 schedule. HFHS-fed rats obtained significantly less rewards during FR schedule compared to the control chow-fed rats (effect of group: $F(1,10)=5.31$: $P<0.05$). Three days after the end of the FR schedule, rats were switched to a PR schedule. Over the last five sessions, rats on the HFHS-choice diet showed significantly more active lever presses, compared to the control chow-fed rats (repeated measures ANOVA: $F(1,10)=10.00$; $P<0.02$), and obtained significantly more rewards ($F(1,10)=9.80$; $P<0.02$). The active lever presses data and reward data of the five sessions were pooled and are shown in Figure 5. The speed of responding per obtained reward was calculated for both HFHS-choice diet and chow-diet rats. The speed was similar for the first nine rewards, thereafter the speed declined for control rats, while the HFHS-choice diet rats kept pressing at similar speed (effect of group: $F(1,10)=5.4$; $P<0.05$; Figure 6).

Food intake in the home cage during the FR schedule was not different from the weeks before the schedule (Table 2), and the amount of sucrose consumed in the home cage did not correlate with the amount of lever presses in the operant cage under the FR schedule (R^2: 0.007; $P=0.8$). During the PR schedule, sucrose solution consumed in the home cage did not correlate with the amount of active lever presses to obtain a sucrose pellet (R^2: 0.02; $P=0.8$). Also, the PR schedule did not influence food intake in the home cage (Table 2). Because we had only six operant cages to test the rats, half of the rats were tested directly after lights went off (at 7.30 h), and half of the rats were tested 4 h in the dark period. To ensure that this difference would not affect the

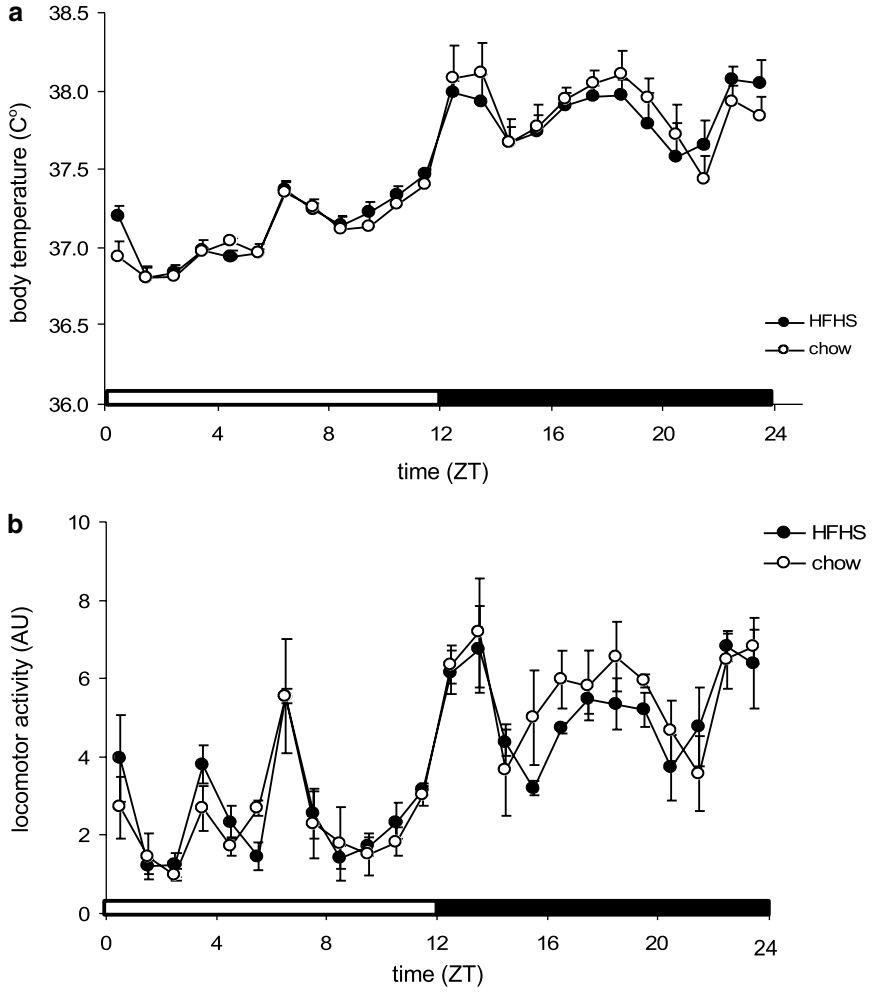

Figure 2 Body temperature (**a**) and locomotor activity (**b**) in HFHS-choice diet and chow-fed rats. Black bars represent the dark period. Animals were handled to measure food and body weight at ZT 6, which increases both body temperature and locomotor activity. Data are mean ± s.e.m. ($n = 4$).

Table 1 Hormone concentrations and organ weights in HFHS-choice diet and control diet rats

	Chow	*HFHS-choice*
Glucose (mmol/l)	5.8 ± 0.1	6.5 ± 0.2*
Insulin (ng/ml)	2.2 ± 0.4	1.9 ± 0.3
Leptin (ng/ml)	3.0 ± 0.2	4.3 ± 0.4*
Thymus weight (mg)	512 ± 61	441 ± 37
Adrenal weight (mg)	45 ± 5	51 ± 5

Abbreviation: HFHS, high fat high sugar. *$P < 0.05$.

animals' behavior, we alternately tested the rats in the first or second session from one day to the next. Furthermore, we tested whether the time of testing was influencing the amount of total lever presses. There was no difference in the amount of active lever presses when tested under a progressive ratio when starting in the morning or 4 h into the dark period (data not shown).

Correlation between food-motivated behavior and obesity

After finishing the progressive ratio schedule, 18 of the 30 chow-fed rats were subjected to the HFHS-choice diet, whereas 12 remained on the chow diet. HFHS-choice diet rats consumed more calories than the control rats eating only chow (Table 3). After four weeks on the HFHS-choice diet, rats were heavier compared to the control rats and had significantly increased fat stores (Table 3). Abdominal fat (abdominal white adipose tissue (AWAT)–epididymal, perirenal and mesenteric) corrected for body weight correlated positively with the amount of active lever presses under the PR schedule only in rats subjected to the HFHS-choice diet (Figure 7a; HFHS-choice diet: $R^2 = 0.5$; $P < 0.0001$, chow diet: $R^2 = 0.1$; $P = 0.3$), whereas subcutaneous fat stores corrected for body weight did not correlate with active lever presses under the PR schedule in either group ($P = 0.2$, data not shown). Average daily calorie intake did not correlate significantly with the active lever presses under the PR

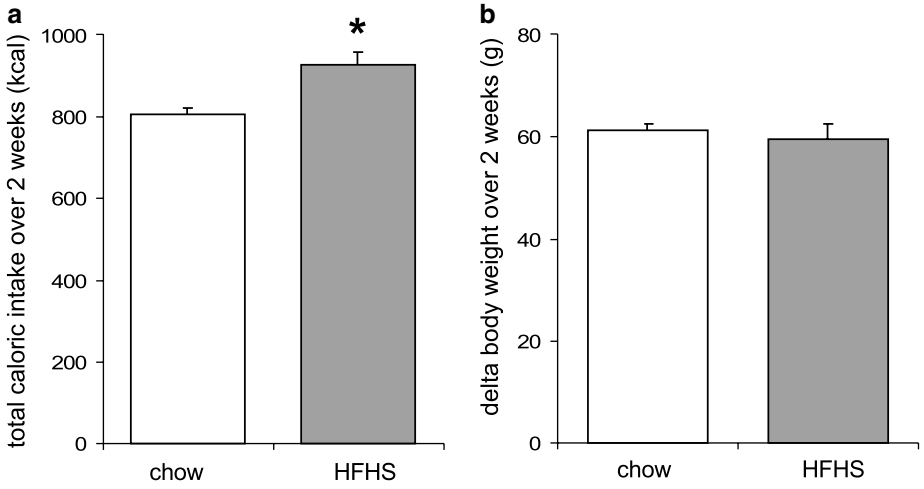

Figure 3 Total calorie intake (**a**) and delta body weight (**b**) over 2-week period when subjected to the chow diet (white bars) or HFHS-choice diet (gray bars). Data are mean ± s.e.m. ($n = 6$).

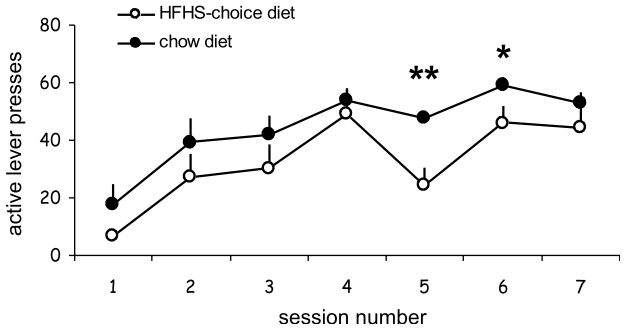

Figure 4 Mean active lever presses of chow-fed and HFHS-choice diet rats per session under the FR1 schedule. Data are mean ± s.e.m. ($n = 6$). * $P < 0.05$; ** $P < 0.01$.

schedule ($R^2 = 0.12$; $P = 0.18$). Other food intake measures (such as percentage lard, sucrose or both) did not correlate with active lever presses.

Discussion

Consistent with previous studies, we showed that the choice diet with a highly palatable lard and a sucrose solution (HFHS-choice diet), in addition to the rats' normal chow, resulted in obesity with increased calorie intake as compared to the chow-fed rats.[16,17] One week on this diet increased abdominal and subcutaneous fat stores and increased plasma concentrations of glucose and leptin. In addition, several weeks on the HFHS-choice diet increased the motivation for food as measured with a PR schedule of reinforcement. Interestingly, we were able to correlate obesity with food-motivated behavior tested before rats were subjected to the HFHS-choice diet. The more motivated the rats were to respond for sucrose pellets, the more abdominally obese they became when subsequently subjected to an obesogenic environment. Because the HFHS-choice diet itself also increased the motivation to press for food, a vicious circle can be established that makes it difficult to fight the motivation to eat when being obese.

Because one week was enough to develop increased adiposity with our HFHS-choice diet, we started testing food-motivated behavior in rats after two weeks on the diet. In this way, fat stores would be increased and the rats would be relatively obese compared to chow-fed controls. We observed an increased motivation to press for sucrose in HFHS-choice diet rats as shown by the increased active lever presses under a PR schedule of reinforcement. This increase was characterized by an increased speed to respond for sucrose during the latter parts of the session, not because of an overall increased rate of pressing. Thus, our HFHS-choice diet rats displayed a prolongation of the motivation to work for sucrose. Moreover, increased responding for sucrose was not due to hyperactivity as the HFHS-choice diet did not change locomotor activity in the home cage. The increased motivation in the obese rats on the HFHS-choice diet is consistent with the reported increase in food-motivated behavior in genetically (Zucker rat) and diet-induced obese animals, as measured by total responses on fixed-ratio and variable-interval schedules.[22,23] However, Glass and colleagues found a difference only between obese and lean Zucker rats when pressing for grain pellets, and not for sucrose pellets.[26] They hypothesized that obese rats are more responsive to the energy content, which is higher in sucrose than in grain pellets. It is, however, important to consider the influence of satiety signals in responding for food. Indeed, when we tested rats under the FR1 schedule with a maximum of 60 sucrose pellets, we observed a decreased amount of total presses in obese HFHS-choice diet rats. Apparently, when rats consume more than 25 pellets, satiety decreases the motivation for food, interfering

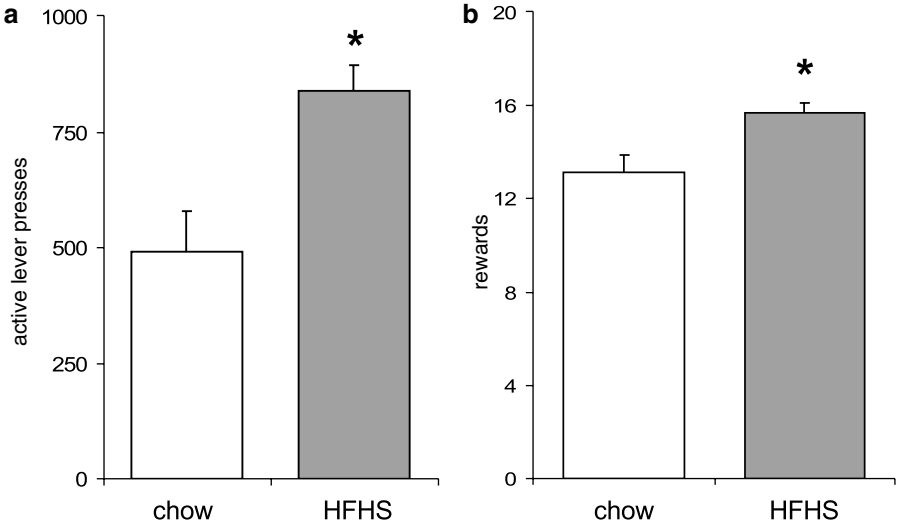

Figure 5 (**a**) Mean active lever presses of chow-fed and HFHS-choice diet rats under the PR schedule. (**b**) Mean rewards obtained in chow-fed and HFHS-choice diet rats. Data are mean ± s.e.m. ($n=6$). *: $P<0.05$.

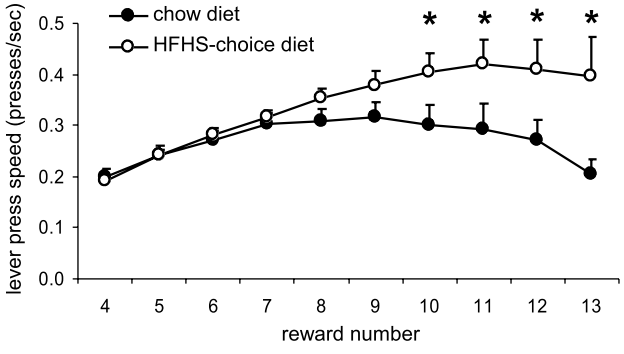

Figure 6 Mean lever press speed per reward obtained under the PR schedule in chow-fed (solid circles) and HFHS-choice diet rats (open circles). Data are means ± s.e.m.($n=6$) over the 5 last days. *: $P<0.05$.

Table 2 Daily total calorie intake in rats before and during FR and PR schedule

	Chow	*HFHS-choice*
Before testing	62.4 ± 2.5	73.3 ± 1.1*
Under FR schedule	64.7 ± 1.0	74.5 ± 1.8*
Under PR schedule	64.9 ± 2.5	75.6 ± 2.6*

Abbreviations: FR, fixed ratio; PR, progressive ratio. *$P<0.05$.

with responding for food. This may also explain the findings by Grass *et al,* because under their PR3 schedule of reinforcement rats obtained over 40 pellets of sucrose.[26] With the PR schedule of reinforcement we used in these studies, we did not observe a satiety effect, and therefore our data suggest that obese rats are more motivated to work for food.

Table 3 Parameters of rats in experiment 3

	Chow	*HFHS-choice*
Average total daily calorie intake (cal)	68 ± 2	92 ± 2**
% chow	100	39 ± 1
% lard		34 ± 2
% sucrose		27 ± 1
Delta BW over 4 weeks (g)	60 ± 2	88 ± 7*
Final fat weight (mg/100 g BW)		
Mesenteric	902 ± 38	1304 ± 74**
Epididymal	806 ± 38	1120 ± 45**
Perirenal	779 ± 22	1219 ± 37**
Subcutaneous	864 ± 22	1227 ± 43**

Abbreviations: BW, body weight; HFHS, high fat high sugar. *$P<0.05$; **$P<0.001$.

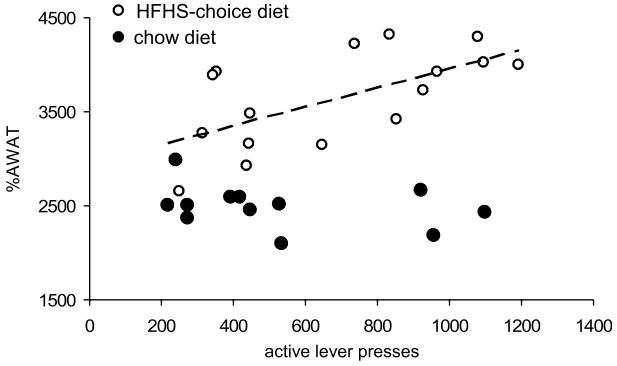

Figure 7 A correlation between active lever presses (mean of last 5 days) and abdominal fat stores as percentage of total body weight (% AWAT) in chow-fed (solid circles) and HFHS-choice diet rats (open circles).

Interestingly, motivation to respond for sucrose before rats were subjected to the HFHS-choice diet correlated positively with the rate of obesity afterwards. This finding was specific for abdominal fat stores. Furthermore, this correlation was also selective for the rats subjected to the HFHS-choice diet, as chow-fed rats did not show this correlation. This suggests that increased motivation for palatable foods provides a critical factor for visceral obesity. Therefore we hypothesize that food-motivated behavior is an important component in the development of obesity when subjected to an obesogenic environment.

Interestingly, only the abdominal fat stores correlated with food-motivated behavior and not the subcutaneous fat stores. Although the underlying mechanisms are unclear, there is a separate organization of the neural innervation of abdominal and subcutaneous fat stores that provides a basis for differential increases in these fat depots.[28] Thus, it could well be that neural mechanisms underlying the increase of abdominal fat stores due to HFHS-choice diet overlap with neural mechanisms underlying food-motivated behavior. However, this hypothesis requires further investigation.

Because food intake is an important determining factor for obesity, it would be logical to assume that calorie intake is directly correlated with the motivation to respond for sucrose. However, the motivation to respond for sucrose did not correlate with future intake of palatable foods (or total calorie intake) when subjected to the HFHS-choice diet. This may be explained by the fact that calorie intake did not vary enough within the group of rats eating the HFHS-choice diet, compared with the variation in active lever presses under the PR schedule. This suggests that food-motivated behavior correlates with abdominal obesity, but that the obesity is not just a consequence of increased total calorie intake. Further studies are necessary to investigate possible additional factors involved.

It has been reported that food restriction increases the motivation to press for sucrose,[29] but food restriction changes the energy status of the rat. Therefore, we chose not to food restrict the rats before measuring motivation. This, however, increased the risk that the time the rats were tested during the dark period (in which they consume most of their food) would influence the data. We show that there were no differences between rats tested 4 h in the dark period (when rats were more satiated) compared to rats tested right after dark onset (when rats are less satiated). In addition, the amount of rewards obtained during testing did not affect calorie intake in the home cage. Therefore, we feel confident that we measured motivation for food, independent of feeding behavior in the home cage.

There are few studies using PR schedules in human subjects.[18,19,30] It has been argued that performance under the PR schedule provides a measure of motivation to obtain specific foods rather then a measured hedonic value ('liking'). For example, alterations in mood affect performance under a PR schedule using chocolate as reinforcer in humans.[30] It is so far unknown, whether obese individuals differentially respond under a PR schedule.

One week of consuming the HFHS-choice diet resulted in obesity, with increased abdominal and subcutaneous fat stores and increased plasma leptin concentrations. Furthermore, body temperature and locomotor activity were not affected by the HFHS-choice diet. Leptin concentrations correlated positively with abdominal fat stores in Experiment one, a finding consistent with earlier findings.[17] It is interesting that obese rats showed increased motivation for sucrose while having increased leptin concentrations. Leptin has been shown to reduce the rewarding effect of lateral hypothalamic self stimulation at food sensitive sites,[31,32] and recently it has been shown that leptin affects mesoaccumbens dopamine signaling.[33,34] This suggests a role for leptin in the modulation of brain reward mechanisms. It would, therefore, be logical to assume that leptin decreases motivation to press for sucrose. Indeed, obese Zucker rats that lack a functional leptin receptor have increased motivation to press for sucrose.[22,23] In view of this, the increased leptin concentrations in our HFHS-choice diet rats would suppress the motivation to obtain a sucrose reward. However, we do observe increased food-motivated behavior. This could be explained by changes in leptin sensitivity in HFHS-choice rats or by a further increase in food-motivated behavior in the absence of leptin alterations. Interestingly, like food-motivated behavior, leptin sensitivity has also been shown to predict abdominal fat gain when rats are subjected to a high-fat diet.[35] Rats that were relatively insensitive to leptin's anorexigenic effect were more prone to obesity when fed a high-fat diet.[36] Because leptin-sensitive neurons are also present in areas important for reward mechanisms,[31,32,37] it would be interesting to study whether the leptin sensitivity and the motivation to press for sucrose that both predict future obesity are related.

Plasma glucose concentrations were increased after 1 week on the HFHS-choice diet. On the other hand, plasma insulin concentrations were not increased, suggesting impaired glucose tolerance. Whether this increased blood glucose is due to altered glucose production or utilization was not examined in this study. It is, however, interesting that a diet with only chow and lard did not result in increased blood glucose concentrations[17] (la Fleur *et al*., unpublished observations). It has been shown that 1 week exposure to both a high-sucrose diet or a high-fat diet results in increased gluconeogenesis, but not in increased basal blood glucose concentrations.[38] There is, however, evidence that a high-sucrose diet changes glucose utilization by changing insulin sensitivity especially in the muscle.[39] Whether our HFHS-choice diet changes glucose utilization remains to be determined.

In summary, we show that the harder a rat is willing to work for sucrose, the more obese it will become when subjected to an obesogenic diet. Instead of proper counter-regulatory mechanisms that should decrease feeding in the light of increased adiposity, the motivation for palatable

sucrose pellets is actually higher in obese rats. Thus already more motivated, obesity makes the rat work even harder for sucrose pellets.

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