What is the Effect of Genotype on High Fat Food Consumption with Different Prices?

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USDA Higher Education Challenge
Grant # 2007-38411-18146

August 14, 2009

Thesis Statement:

High fat food consumption patterns vary according to the price of the food as well as between different genotypes of mice.

Abstract:

Using female mice as subjects, the following accounts for an economic behavioral study to address genetic differences in consumer demand of high fat food as the price of the food increased. Mice were place into a steel operant chamber and trained to press a lever to procure high fat food (35% fat). Lever presses were used to simulate price and the price of the food increased each week for four weeks total. Behavioral observations were made as the price of the food increased, and reward, weight, glucose tolerance and estrous cycle measurements were recorded. Mice of the genotypes TubMUT and wild-type were used to see if food consumption patterns differ among genotypes. The experiments show that consumption patterns in relation to price do differ among female mice of different genotypes. Though the economic law of demand states that consumption of the high fat food should decrease as its price increases, not all mice demonstrated this law in their behavioral patterns. Additionally, it is shown that a relationship between female mouse estrous cycle stage and volume of food consumption may exist. Further research will be necessary to determine the strength and validity of the relationship. Overall, this study shows that, if applied to humans, genetic specific approaches to food taxation may be necessary in order to effectively deter people from consuming large quantities of high fat food.

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Introduction:

The obesity epidemic in America is becoming more severe each year. Between 1960 and 2003, the percentage of obese and overweight Americans almost tripled from 13% to 34% [1]. During this time, fast food restaurants and convenience stores have become ubiquitous across the country, and advancing technology and agricultural practices have allowed for foods to be cheaper and more available to the average consumer [2]. This increasing food availability has lead to the trend of high sugar and high fat foods being cheap, readily available and easily consumed by the average person, regardless of socioeconomic status [3, 4]. Between 1970 and 2003, the consumption of fats and oils increased by 63%, grains by 43%, and that of sugars by 19%, while the average fruit and vegetable consumption only increased by 12% and 24%, respectively [5]. Also since 1970, the per capita daily intake has increased by 523 calories per person, per day to an average of 2,757 total calories [5]. Both have served as possible contributing factors to the growing obesity epidemic [3].

The fact that high fat and high sugar foods are less expensive and more prevalent than fruits and vegetables has led to higher obesity rates, especially among individuals of lower socioeconomic status [4, 6]. Consumer food choices are controlled by taste, cost and convenience and, in situations of deprivation, the human preference for energy-dense high fat and high sugar food functions as an advantageous mechanism for survival [2, 4]. In recent years there have been numerous government efforts to promote decreased consumption of fats and sweets and increased consumption of whole foods. However, due to the inverse relationship between energy density and energy cost, the advice to consume less energy-dense foods results in unintended, but significant, financial strain on consumers [4]. As a result, food prices continue to serve as an obstacle preventing long-term nutritional changes.

Many studies have been conducted to measure the effect of food price on the consumption patterns of mice. These studies have shown that consumption rates and body weight measurements decline as a particular food becomes more expensive and harder to procure [2, 7]. Mice, when given two food options of varying prices, are more likely to eat a greater amount of the lower priced commodity [2, 7-9]. When mice are given only one commodity to choose from, consumption of this commodity decreases as its price increases [7-9]. These trends follow the economic law of demand, which states that as the price of a commodity increases, consumption of that commodity will decrease [2]. Relating these findings back to the trend that food price decreases with increasing energy-density, it is clear to see why being of lower socioeconomic status fosters the consumption of an unhealthy diet and leads to higher risk of obesity [4].

Genetic factors also serve as a factor in the rising obesity rates. In humans and mice, some obese phenotypes are caused more by genetic mutations than the environment. For example, leptin-deficiency is caused by mutations in the gene encoding for leptin, and low serum leptin is linked to obesity in both humans and mice [10]. Another example would be the Tubby mutant mouse model, in which a point mutation in the *Tub* gene is linked to causing an obese phenotype without hyperphagia [11]. This genetic mutation has been linked to increased obesity prevalence due to errors in metabolism and reduced motivation to exercise, but not due to overeating [11].

Our country is moving toward using taxation and subsidies to remove the cost discrepancies between energy-dense foods and nutrient-dense foods to promote healthier eating [4]. However, the United States is a melting pot of individuals of differing genetic backgrounds. As with mice, there is currently little research to show whether individuals of different genetic backgrounds make different food choices while facing similar economic conditions. Gaining a more thorough understanding of the relationship between genetics and economic food choices is essential to making more effective economic policy changes, as well as creating genetic-specific treatment plans for obesity and related diseases.

Specific Aims

This research project is designed to address the relationship between genetic factors, food price and consumption, specifically relating to the consumption of high fat food. The project is designed to test whether increasing the price of high fat foods will decrease its consumption, and to test whether different genotypes of mice react differently to the changing food prices.

Hypothesis

Female mice, regardless of genotype, will decrease their consumption of high-fat mouse food as the price of the food increases. Additionally, price will have less of an impact on tub-mutant mice over their wild-type counterparts as the tub-mutants will be more motivated to obtain the high fat food.

Materials and Methods:

Subjects

Female mice of the genotypes *Tub* MUT (n=1) and wild-type (n=1), ages eight to ten weeks at the start if the study, were utilized. *Tub* MUT mice of the C57Bl/6 mouse line are an obese mouse model and typically show an obese phenotype by the adult age of 9-12 weeks [11]. These mice possess a point mutation in the *Tub* gene that results in adult onset obesity without hyperphagia (overeating), indicating that the mutation causes some sort of error in energy-expenditure or energy-usage within the body [11]. On the contrary, wild-types of the 129Sv/J C57Bl/6 mixed mouse line possess no genetic mutations, thus do not exemplify an obese phenotype with normal food consumption and served as a control for this study [12]. These two different mice models were used to measure whether genetics has an effect in how mice react to the changing high fat food prices.

Operant Chamber Protocol & Vivarium Conditions

Mice chosen based on genotype, age, and sex were first weighted, and then placed into individually housed shoe-box cages for approximately two days and fed 5.0g/day of 20mg-high fat pellets (35% total calories from fat). They were then placed into a steel dual lever operant chamber (Figure 1) controlled by a computer. Mice remained in the chamber 24 hours per day with nestlets and sheppard shacks except during procedures, which took approximately two minutes. Trays were cleaned daily with isopropyl alcohol and water. One side of the cage featured two levers connected to pellet dispensers and the other provided water ad lib. Above each pellet dispenser cup, small cue lights were used to indicate when the levers were active each night. Once trained, mice pressed the levers a specific number of times to receive one pellet of food. The computer and one lever were active from 5pm-5am each night, as this is when mice normally are awake and eat. Conditions of the vivarium were as follows: 70^{o}F, 40% humidity and lights on from 5am to 5pm daily.

Price Structure

Shaping methods, through the use of shaping buttons and peanut butter, was used to train the mice to use levers to obtain pellets. Once a mouse had been proven comfortable with the levers by consuming at least 100 pellets per night at a price of seven presses per pellet, or after about one week, she was then placed on the first price schedule of ten presses per pellet. The price of each pellet was then increase every seven days for four consecutive weeks (Table 1). The prices each week were as follows: 10 presses for schedule one, 23 presses for schedule two, 32 presses for schedule three, and 40 presses for schedule four. Only one lever, either the left or right, was active for the entire course of the study for each mouse.

Measurements

Weight was measured as each mouse began the first price schedule. Throughout the remaining experimental time, mice were monitored daily and their weight, lever presses and reward were recorded. Daily vaginal swabs were administered using 20µL of Phosphate Buffered Saline (PBS) to track stages of the female estrous cycle. At the end of the fourth price schedule, each mouse was removed from the study and final measurements were taken including weight and body fat percent via Magnetic Resonance Imaging (MRI). A glucose tolerance test was also conducted using a standard glucometer. All procedures were done in accordance with the Institutional Animal Care and Use Committee and Institutional Review Board at Virginia Tech.

Data Analysis

At the completion of the study for each mouse, computer data was analyzed for average pellet consumption per night, lever presses per night and average weight per price schedule. Average reward and average weight were plotted against price schedule. Additionally, stages of the estrous cycle were plotted against nightly reward to identify any relationship that may exist between the female mouse estrous cycle and food consumption patterns.

Results

By August 1, 2009 two mice had completely finished the study and their results are discussed here. Overall the results varied greatly between the two subjects. The average reward, average weight and food consumption as related to estrous cycle stage of each mouse, a wild-type female and *Tub* Mut female, are summarized by figures two, three and four.

Food Consumption

The average reward per price schedule (Figure 2) was calculated by dividing the total number of lever presses per night by the price per pellet. The wild-type female's consumption steadily decreased across the three price schedules completed: 139 pellets/night (price=10), 116 pellets/night (price=23) and 36.8 pellets/night (price=32). The total decrease in average reward was 102.2 pellets/night over the three price schedules. The average reward for the *Tub* MUT varied throughout the four price schedules: 122 pellets/night (price=10), 136 pellets/night (price=23), 96.4 pellets/night (price=32), 122 pellets/night (price=40). Two-tailed t-test analysis indicates P=0.558.

Weight

The weight of the wild-type female fell from 21.4g to 16.6g between price schedules one and three, a total loss of 4.8g or 22% of her initial body weight. Meanwhile the *Tub* MUT's weight fluctuated from 19.2g at the start of the study to a max of 20.4g during the third price schedule, then back down to 20.2g at the end of the study. Maximum weight gain of the *Tub* MUT was 0.9g, or 4.6% of her initial body weight, between the beginning and end of the study. Two-tailed t-test analysis indicates P=0.988.

Estrous Cycle Data

Over the course of the study, vaginal swabs were used to tract the female mouse estrous cycle and identify a possible relationship to consumption patterns. Figure 4 graphs the average reward over all four price schedules for each stage of the estrous cycle. The wild-type female consumed an average of 82 pellets/night on days of early estrous, estrous, and post-estrous stages of the cycle, while consuming an average of 109 pellets/night on days of diestrous and proestrous. The Tub MUT female consumed on average 107 pellets/night on days of early estrous, but averaged 121 pellets/night throughout the other four stages of the cycle. Two-tailed t-test analysis indicates P=0.015.

Discussion

Analysis of data

There is a significant difference between the consumption patterns and average weight of the mice of different genotypes. The wild-type female exhibited a negative correlation between food consumption and price as hypothesized. As the price of the high fat food increased, her consumption steadily decreased, exemplifying the economic law of demand [2]. However, the *Tub* MUT female reacted inversely to what was expected by maintaining her consumption as the price of food increased. Also, while the wild-type female lost weight with increasing price, the *Tub* MUT maintained or gained weight over the course of the four price schedules. It was quite concerning that the wild-type female suddenly stopped pressing the lever during the third price schedule, even though her performance beforehand was solid. It is hyothesized that, since working with a female mouse of small initial weight, the small weight loss from one night of low consumption was detremential enough to negatively effect her performance on subsequent nights. This lead to a downward spiral that eventually led to her removal from the study due to weight loss of greater than 20%.

The *Tub* MUT female consumed a consistent level of the high-fat food and gained weight despite rising food costs over the four price schedules. This indicates that increasing the price of the food was an ineffective mechanism to decrease high-fat food consumption and prevent weight gain in this mouse model. Meanwhile, since the wild-type female decreased her consumption with increasing food price, price served as a successful inhibitor to her consumption of the high fat food. The differences in behaviors across the genotypes show that, if applied to humans, food taxes may be effective in decreasing high-fat food consumption in certain genotypes of people and ineffective in others.

Due to the fact that only two mice have fully completed the study, statistical significance is low. A two-tailed T-test showed that P=0.588 and P=0.988 for Figures two and three, indicating that differences in food consumption and weight between two genotypes are not statistically significant.. Although differences between the two genotypes seem significant, more subjects will be necessary to increase the statistical significance of values obtained.

Questions Raised

Previous studies have shown that *Tub* MUT mice possess this obese phenotype while eating less than their wild-type counterparts, attributing the obesity to errors in metabolism and reduced motivation to exercise [11]. In this study the *Tub* MUT's consumption was significantly greater over the four price schedules than the wild-type's consumption, which is contrary to previous literature. It is unclear why the *Tub* MUT showed greater consumption than the wild-type female. Was the *Tub* MUT's consumption pattern due a greater level of motivation over the wild-type to obtain the high-fat food? This question can be more clearly answered once a larger sample size is obtained and through the use of control studies to determine normal food consumption levels among *Tub* MUT and wild-type females.

A confounding variable is the extent to which caloric expenditure from pressing the lever affects the weight of the mice. In this study it is assumed that exercise exertion in pressing the lever does not have a significant effect on weight, yet future research trials may need to be formulated to further investigate this question. Additionally, it is still unclear whether a strong relationship exists between the female mouse estrous cycle and food consumption patterns. A significant decrease in consumption was seen on days of early estrous for both the Tub MUT and wild-type female, which may be related to higher levels of the estrogen hormone on these days. Higher levels of estrogen in female mice is linked to a greater desire to mate, so it can be hypothesized that the females were more interested in finding a mate on these days rather than eating. However, due to the small sample size, it is difficult to make any conclusions as to whether the consumption patterns are due to affects of the cycle or just a coincidence. More data obtained from future mice in this experiment will hopefully give greater evidence of the strength of this relationship.

Future Research and Implications

This study will continue into the fall 2009 semester with the goal of obtaining data from four mice total of each of the two genotypes tub-mutant and wild-type. Additionally, a third genotype, N2 Knock-Out, will be added to the study once mice of this genotype become available. The study will be fully completed with hopes of future application to human studies, and eventually, economic policies. If the mice are more likely to decrease consumption with increasing prices of the high fat food, and if different genotypes of mice respond differently to price changes, then a supplemental study can be conducted to see if these relationships hold true for humans [2]. An economic approach to fighting the obesity epidemic is one that our country is just beginning to address and could be the exact approach our country needs [3]. Increasing the price of high fat foods can serve to deter more people from consuming such high quantities of it [2, 3]. A supplemental decrease in price or subsidizing of whole foods, namely fruits and vegetables, can hopefully change food consumption trends in America and turn around obesity trends [3]. Furthermore, from this research human trials can be initiated to see if genetic differences affect food choice under various economic conditions. If differences do exist, this would establish and support the need for a genetic specific approach to fighting obesity in our society. With obesity rates on the rise, our country is in critical need of an innovative and effective intervention to reverse the trend and in turn, build healthier lifestyles.

Figure 1: Operant Chamber Design

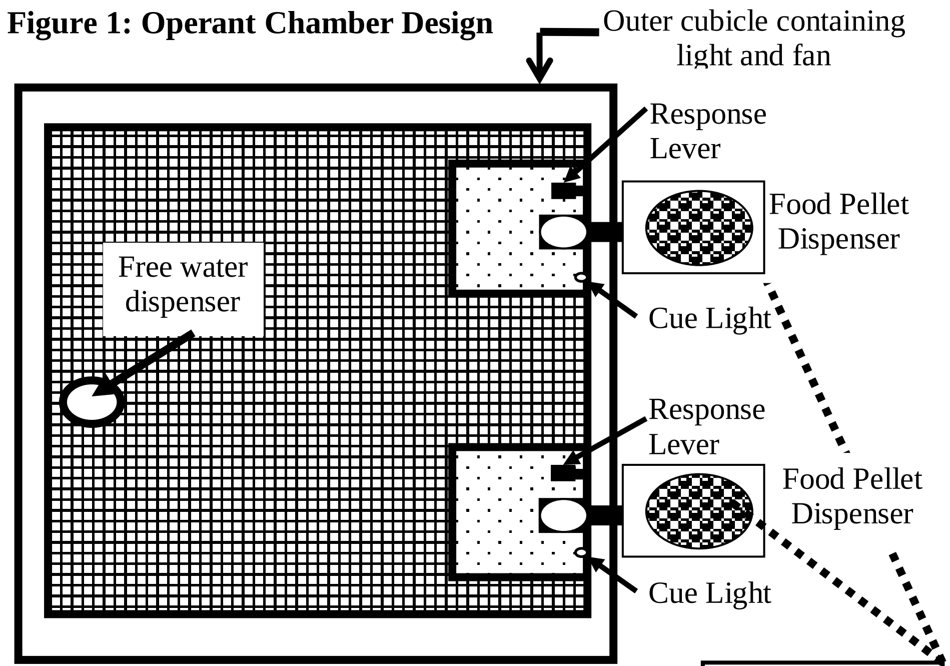
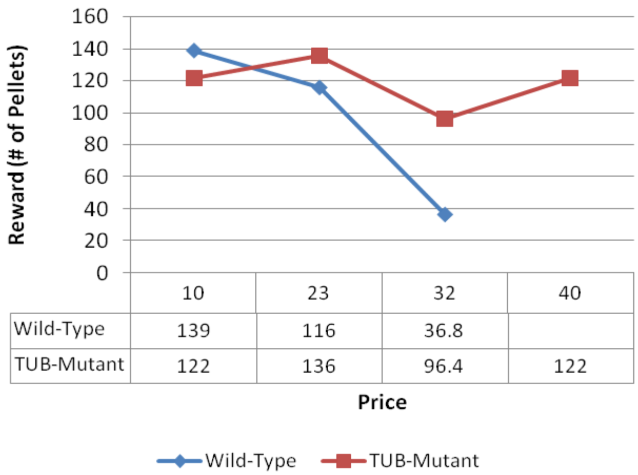


Fig 1. Dual dispenser cage for proposed experiments (Lafayette Instruments). Edited by K.DiCarlo.

Table 1: Pricing Schedule

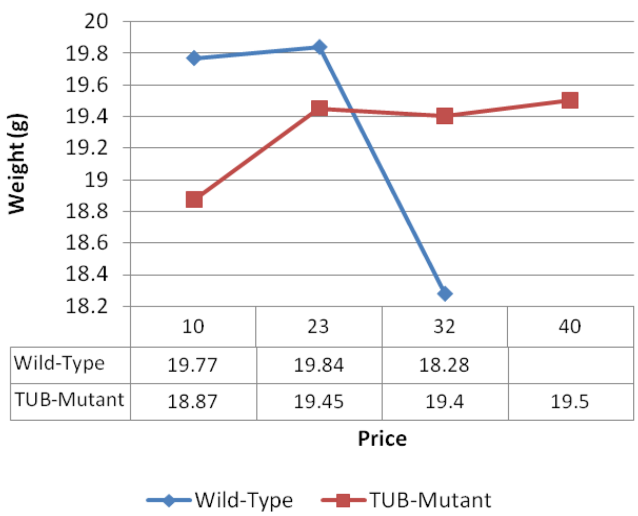
Price Schedule	Price (No. of Presses)	No. Of Days	No. of Days in Dataset
Shaping	1/2/5/7	5-7 days	0
1	10	7	Last 5
2	23	7	
3	32	7	
4	40	7	

Figure 2:
Reward v. Price Schedule



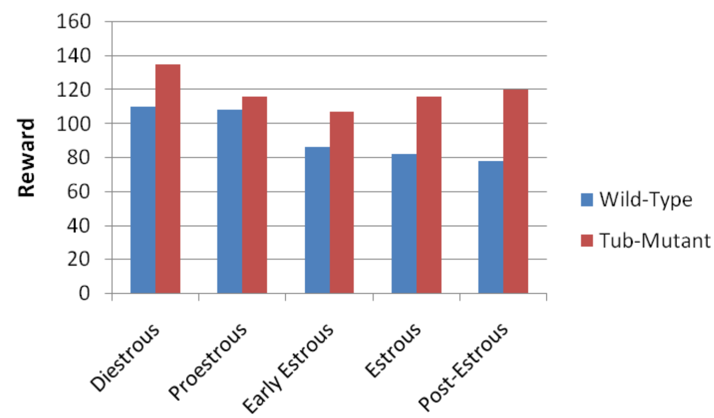
	10	23	32	40
Wild-Type	139	116	36.8	
TUB-Mutant	122	136	96.4	122

Figure 3:
Average Weight Change v. Price Schedule



	10	23	32	40
Wild-Type	19.77	19.84	18.28	
TUB-Mutant	18.87	19.45	19.4	19.5

Figure 4:
Reward vs. Estrous Cycle Stage



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