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CONSUMPTION OF A HIGH FAT DIET IN ADOLESCENT RATS:
THE EFFECTS ON ADULT FOOD PREFERENCES

By

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A Thesis Submitted in Partial Fulfillment
Of the Requirements for a Degree with Honors
(Dietetics)

The College of Health and Human Services

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Introduction:

Childhood obesity is a growing epidemic in the United States. According to the Centers for Disease Control and Prevention (CDC), it is estimated that 17 percent of children between the ages of 2 and 19 are obese and this number has been on the rise¹. It is important to research the basis for why children are becoming obese and how obesity can affect them when they reach adulthood. The CDC states that overweight and obese children and adolescents are more likely to develop risk factors associated with cardiovascular disease such as high blood pressure, high cholesterol, and type 2 diabetes, than children who are not overweight or obese¹. There have been studies that suggest that many children who are overweight or obese during adolescence continue to be overweight or obese when they are adults^{2,3}.

Several studies have been conducted in attempt to answer the general question of what behaviors or diets in childhood or adolescence can lead to obesity and other co-morbidities that can lead to chronic diseases in adulthood. One study examined maternal diet during gestation and lactation of rats and how it related to dietary behaviors of offspring, particularly in the consumption of high fat foods⁴. The researchers found that the mother rats, which were fed a 'junk food' diet during gestation and lactation, had offspring that were more likely to over consume high fat, sugary and salty foods at the expense of protein rich foods⁴. This was in comparison to the offspring of mothers that were fed a chow diet.

In another study, researchers examined the short-term effects of feeding juvenile rats a high fat diet compared to a low fat diet and their fat preferences after four weeks⁵. In

that study, one group of female rats was fed a 10% fat diet and the other group of female rats was fed a 30% fat diet. The researchers tested fat preference with three different peanut butter based mixtures. During testing periods the rats had access to the mixtures for two hours. Data collected from this study showed that juvenile rats fed a high-fat diet consume more total fat during short-term tests relative to animals fed a diet lower in fat⁵.

The aim of the present study was to determine whether there are long-term effects of consuming a high fat diet during adolescence on food preferences later in life. In order to determine effects in the long-term, the rats were given access to a low-fat diet after being maintained on their designated diets throughout adolescence, postnatal day 24 to postnatal day 84. In adulthood, two preference tests were performed three weeks apart. We hypothesized that rats maintained on a high fat diet throughout adolescence would show an increased preference for highly palatable foods in adulthood by consuming more grams of a palatable food, sweetened vegetable shortening, during the preference test.

Methods:

In order to conduct this study, 25 male Long Evan rats (Harlan, Indianapolis, IN) were weight matched into 3 different groups. The groups were chow, high fat and pair fed rats. The three groups contained 8 rats, 8 rats, and 9 rats respectively. Each group was individually housed in stainless steel hanging wire cages with ad libitum access to water. The room was temperature and humidity controlled with a 12:12 hour light/dark cycle. Food intake was determined daily and spillage was collected and accounted for in the weight calculations. Rats were weighed daily. Group 1 was designated the chow group and had ad libitum access to a chow diet throughout adolescence. Group 2 was designated the

high fat group and had ad libitum access to a high fat diet throughout adolescence. Group 3 was designated the pair fed group and was fed a high fat diet that was calorie matched to the intake of the chow group. Each day, the average calorie intake for the chow group was calculated and then the amount of grams of high fat diet was calculated to match these calories and given to the 9 pair fed rats.

Food hoppers were used for the pellet food. Spill papers (e.g. scrap computer paper) were used under each individual cage and changed daily. Water bottles were single-holed stopper and a stainless steel drinking spout was attached to each cage with a wire attachment spring. All procedures were approved by the Purdue University Animal Care and Use Committee.

Each group was 24 days old when they were started on each of the designated diets and remained on these diets until study day 60. It is from weaning to postnatal day 60 that rats are considered adolescent⁶. On study day 60, postnatal day 84, each group was switched to a low fat diet. The macronutrient content of each diet is listed in Table 1. The groups remained on the low fat diet for 14 days. Throughout these 14 days, the sweetened vegetable shortening (SVS) was offered at 3 random times in order to prevent neophobia during the preference testing cycle. In previous studies, SVS has been used successfully as palatable binge food^{7, 8}. On study day 74, the first preference test was performed. After the first preference test, each group was then placed back on the low fat diet for 21 more days and then on day 95 the second preference test was performed. Weight, food intake, and spill were recorded daily for each rat. NMR measured body composition of each rat once a week.

Preference testing was performed over a 12-hour period. Each test was started at the onset of the dark cycle and concluded at the end of the dark cycle. All groups were offered two dishes, one containing the low fat diet and the other containing SVS. Water was offered ad lib throughout the test. The food was weighed before and after the test. Also, spill and weights for each rat were recorded. This style of preference testing is a model from a previous study conducted by Martin and Mullen in order to test macronutrient diet selection behavior in rats⁹.

Table 1: Diet Compositions

Diets	%kcal Protein	%kcal CHO	%kcal Fat	Total kcal/gm
Chow Diet (C)	28	60	12	3.1
High Fat Diet (HF)	20	35	45	4.73
Low Fat Diet (LF)	20	70	10	3.85
*Diets ordered from Research Diets Inc.				
*Sweetened Vegetable Shortening (SVS) is 90% vegetable shortening and 10% sugar that will be mixed in the lab.				

At the conclusion of the study the rats were euthanized by an overdose of sodium pentobarbital (0.4ml/rat providing 390 mg/kg of sodium pentobarbital administered i.p.).

Statistical Analyses

Data are shown as mean \pm SEM. Data were analyzed using PRISM. All data were analyzed using analysis of variance (ANOVA) and Tukey's post-hoc comparisons were utilized to determine differences among groups. A p-value of ≤ 0.05 was accepted as significant.

Results:

The results of the preferences testing did not show a statistically significant difference in any of the groups between their intake in calories of the low fat diet or SVS (Figure 1 and 2). Also, there were no significant differences in the intakes of either diet between the 3 groups. However, over the 12-hour preference-testing period there were significant changes in weight among the groups. There was a statistically significant difference between the pair fed group and the chow group and also between the pair fed group and the high fat group ($P = 0.0096$). The recorded weight changes are depicted in Figure 4.

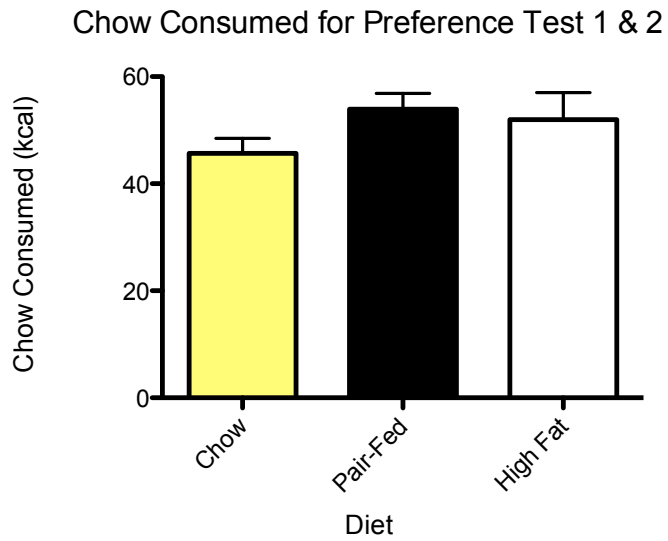


Figure 1: The average intake of kcals from the low-fat chow diet for both preference tests.

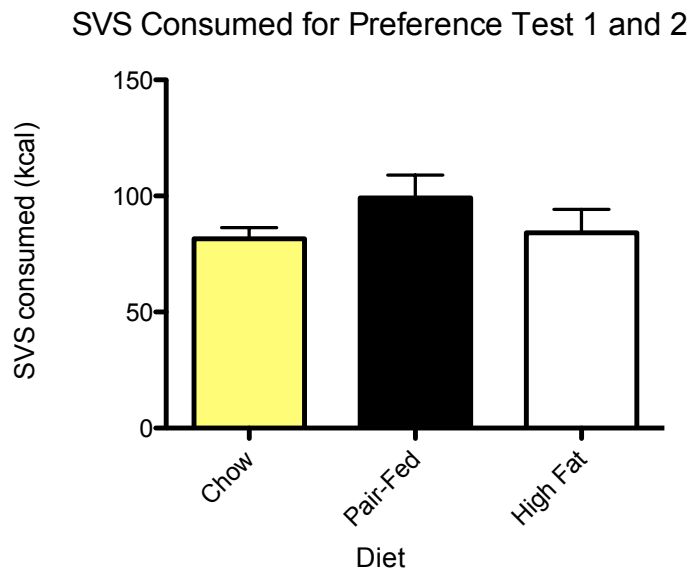


Figure 2: The average intake of kcals from the SVS for both preference tests.

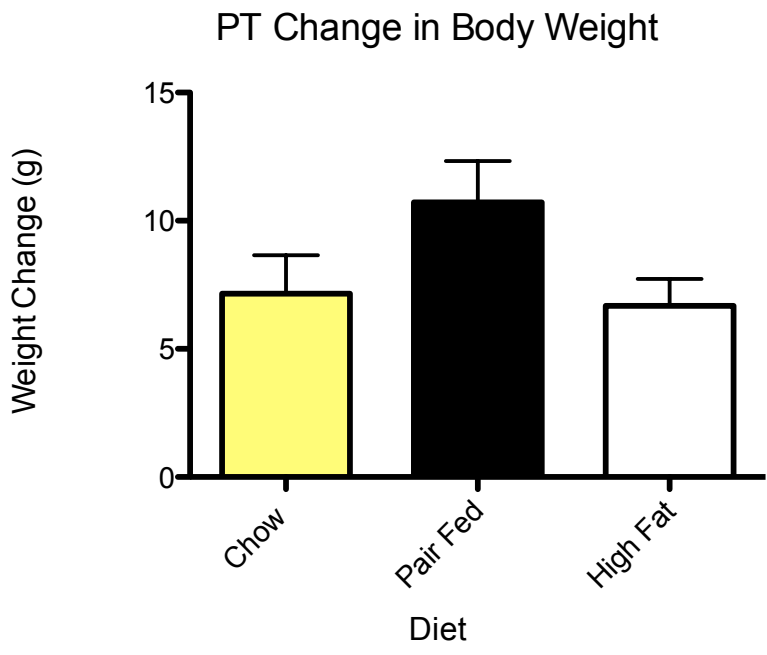


Figure 3: Weight changes for each group during the 12-hour preference test 1.

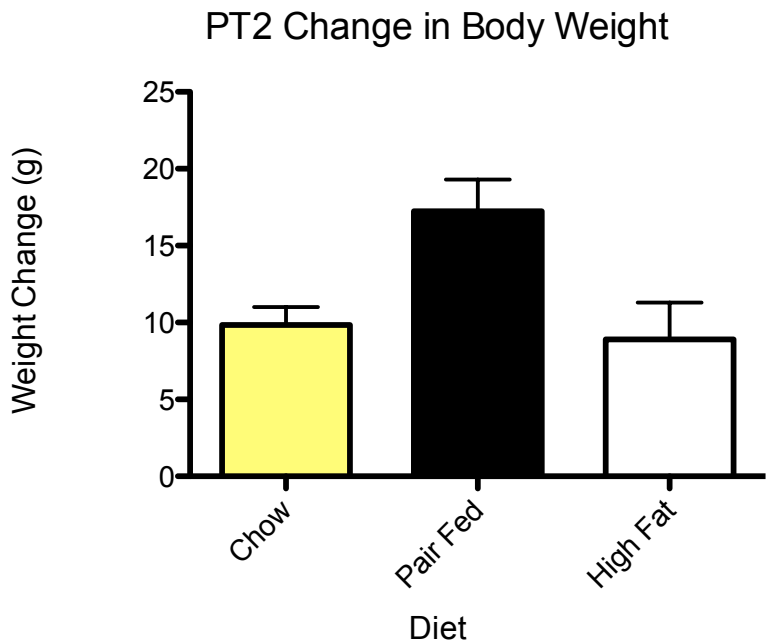


Figure 4: Weight changes for each group during the 12-hour preference test 2.

Conclusion and Discussion:

The result of this study demonstrated that there was no statistical relation between the diet consumed in adolescence and food preferences in adulthood. However, during the 12-hour preference-testing period, there was a significant difference in weight gain in the pair fed group when compared to the chow or high fat diet group. The weight changes are shown in Figure 4. Although there was no statistical significance in kilocalorie intake, Figure 1 and 2 depict that the pair fed group did take in more kilocalories than the other two groups. This may be due to the fact that the pair fed group had a significant amount of weight gain over that 12-hour period and were more affected by the kilocalorie intake. The average intake of kilocalories from SVS was greater than the average intake of the low fat chow diet. If the test groups were larger, significant differences in intake preferences may be seen.

The significant weight gain in the pair-fed group differs from the original hypothesis that the group with ad libitum high fat food during adolescence would eat more kilocalories, which should lead to a greater weight gain. However this was not the case in this study, raising the question as to why statistically significant weight gain occurred in the pair-fed group. It could be possible that the pair fed rats got used to eating a set amount of food and when they were presented with ad libitum food, they over consumed the food offered. This over consumption, compared to their normal consumption could have resulted in more fat storage during the testing period and weight gain. This may be a finding to research further.

In the study by Bayol et al, researchers determined that a maternal junk food diet before weaning promotes an exacerbated preference for junk food and leads to a greater propensity for obesity in the offspring⁴. Results of the current study did show it might be possible that the rats exposed to a high fat diet throughout adolescence (the pair fed group) are at a greater risk for weight gain as shown by the significant weight gain over the 12-hour testing period. However, the kilocalorie intake consumed over the preference test does not support the amount of weight gained shown or the theory that consumption of a high fat diet throughout adolescence can lead to a greater risk for obesity in adulthood.

In the same study, it was shown that young offspring, along with the pregnant and lactating dams exhibited a preference for palatable foods⁴. These were foods that are rich high in fat, sugar, and salt over nutritionally balanced chow⁴. Those findings were not replicated in the current study.

Another study examined at the effect of juvenile consumption of a high fat diet in females and in the short term⁵. In that study the researchers compared intake of total grams of fat between a group with access to a 30% fat diet and a group with access to a 10% fat diet. This study concluded that in the short-term (4 week period), the rats fed a high fat diet consumed more total fat than those fed a low fat diet⁵. In the current study, a similar method was used but we found no significant difference in fat intake or preference. However, we examined the effects of a high fat diet in adolescence in more of a long-term setting. The rats were tested on postnatal day 98 and postnatal day 126, while the rats from the short-term study were tested four weeks postweaning⁵. One of the other major differences in the short-term study and the current study are the high fat mixtures

containing peanut butter, that were offered during testing periods. If the study were done with more similar mixtures, more similar results may have been found.

It is important to look further into the long-term effects of what a high fat diet in adolescence can have in adulthood. Early nutrition could be a major contributing factor to adult obesity and the chronic diseases that can follow. It could be possible to examine other diets, such as various levels of fat content and different forms of fatty foods, that can have long term effects on adolescents. In addition, public prevention and education on obesity may be key to reducing the prevalence of obesity in children. In order to prevent childhood obesity, research must further elucidate contributing factors to the onset and maintenance of overweight and obesity.

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