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FAT-PAD SPECIFIC EFFECTS OF LIPECTOMY ON APPETITIVE AND CONSUMMATORY INGESTIVE BEHAVIORS IN SIBERIAN HAMSTERS (*Phodopus*

sungorus)

by

KELLY DESHON JOHNSON

Under the Direction of Timothy J. Bartness

The aim of this experiment was to test whether lipectomy (LIPX)-induced decreases in body fat affect appetitive (foraging effort and food hoarding) or consummatory (food intake) ingestive behaviors and whether the effects of LIPX on these behaviors is affected by changes in energy expenditure produced by varying the amount of work required to obtain food. This was accomplished by housing male Siberian hamsters (*Phodopus sungorus*) in a foraging/hoarding apparatus where food pellets (75 mg) could be earned by completing various wheel-running requirements. Requiring a foraging effort (10 revolutions/pellet) abolished the normal compensation of WAT mass by the non-excised WAT pads that typically follows IWATx or EWATx. After foraging, food hoarding was increased more than food intake when hamsters were required to forage for food (10 revolutions/pellet). The magnitude of the LIPX-induced lipid deficit (IWATx > EWATx) did not correspond to a proportional change in either appetitive or consummatory ingestive behaviors.

Index Words: obesity, consummatory, appetitive, foraging, food hoarding, food intake

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An Honors Thesis Submitted in Partial Fulfillment of the

Requirements for Graduation with Undergraduate Research Honors

in the College of Arts and Sciences

Georgia State University

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Introduction

Obesity is a result from an energy imbalance where there is an excess body weight in the form of fat. Although everyone needs a certain amount of body fat for stored energy, heat insulation and shock adsorption, the problem is people store energy in far excess of what is needed for homeostasis. This is the result of energy intake (eating too many calories) chronically exceeding energy output (getting enough physical activity). The answer to the obesity problem is to increase energy output and decrease energy input; however, people do not like this answer, as it is very effortful. One strategy to combat obesity is to determine the physiological mechanisms underlying energy intake and expenditure with the hope that this knowledge might lead to less effortful means of decreasing body fat such as development of a pharmaceutical to decrease food intake and/or increase energy expenditure.

The ingestive behavior of non-human and human models consists of both appetitive and consummatory phases and it has been shown that both ingestive behaviors contribute to the overall energy strategy of animals. The appetitive phase is foraging, transporting and storing the food, whereas the consummatory phase is the actual eating of the food. The mechanisms underlying consummatory ingestive behaviors have received considerable attention compared with those underlying appetitive ingestive behaviors, even though both phases are involved in a variety of species. The behavioral response after food deprivation includes changes in both appetitive and consummatory phases in various species, including Siberian hamsters. The behavioral response of Siberian hamsters (Phodopus sungorus) after a fast is to increase two appetitive behaviors, foraging and food hoarding more than food intake (Day, 2003). Other animals, such as laboratory rats, also show an increase in food hoarding, but food intake as well.

Unlike laboratory rats, hamsters are natural hoarders equipped with special anatomical structures, cheek pouches, that permit studies designed to study food intake and food hoarding in a more naturalistic environment. Therefore, Siberian hamsters offer an ideal model to study the differential expression and underlying mechanisms of appetitive and consummatory ingestive behaviors. I propose to use a semi-natural model of foraging and hoarding because both contribute to ingestive behaviors and increasing foraging effort alone alters energetic strategy of animals; specifically, requiring hamsters to run 10 revolutions per pellet, increases food hoarding more than food intake (Day and Bartness, 2001). A combination of the foraging system developed by Perrigo and Bronson and the hoarding system developed by Bartness will be used. Perrigo and Bronson (Perrigo and Bronson, 1985) first developed a foraging apparatus using a wheel- running based delivery of food pellets to the effects of various environmental and physiological challenges on foraging behavior. Bartness and Clein (Bartness and Clein, 1994) developed a simulated burrow system to study hoarding. The modified foraging/hoarding system has been previously used in our lab to study behavioral changes (Day and Bartness, 2001).

One might envision that there is an overall regulation of total body fat and that within this hypothetical body fat regulatory system, both appetitive and consummatory ingestive behaviors might be affected by, and might affect, foraging/food hoarding and food intake. In order to directly manipulate total body fat experimentally specific white adipose tissue (WAT) pads can be surgically removed (lipectomy [LIPX]). Previous studies have shown that lipectomy increases food hoarding in Siberian hamsters (Woods, 1997). It has also been shown in Siberian hamsters that after LIPX, non-excised fat pads are increased to compensate for the lost lipid (Mauer and Bartness, 1997). The aim of this experiment was to test whether LIPX-induced

decreases in body fat affect appetitive (foraging and food hoarding) or consummatory (food intake) ingestive behaviors and whether the effects of LIPX on these behaviors is in turn affected by changes in energy expenditure produced by varying the amount of work required to obtain food.

Materials and Methods

Animals and Housing

The experiment was done in two replicates, with sixty-eight male Siberian hamsters (*Phodopus sungorus*) (~3 months of age) housed individually. The foraging/hoarding system, described previously, was used and was developed in Dr. Timothy Bartness's laboratory (Day and Bartness, 2001). In brief, two cages were connected with a convoluted polyvinylchloride tubing system. The top, food cage contained a water bottle and a running wheel. The bottom cage, contained bedding and cotton nesting material, was covered to mimic a natural burrow. *Procedure*

The hamsters were divided into three different foraging groups: 10 Revolution (10 Revs per 75 mg food pellet, n=24); Free Wheel (FW, exercise, non-foraging control group, n=22); Blocked Wheel (BW, sedentary, non-foraging control group, n=20.) In the 10 Revolution group, the wheel revolutions were monitored using a magnetic detection system and monitored by a computer-based hardware-software system, which also released the pellets. Revolutions were monitored, but do not cause the release of pellets for the Free Wheel group; therefore, food was freely available to these animals (400 pellets). The animals in the Blocked Wheel group also had freely available (400 pellets); the running wheel was immobile.

Before obtaining behavioral measures foraging, food intake and food hoarding, various fat pads were removed to create differential LIPX-induced lipid deficits. These fat pads include epididymal fat (EWAT) and inguinal fat (IWAT). A subset from each foraging group was divided into four LIPX conditions: EWATx + Sham IWAT LIPX (10 Rev n=10, FW n=10, BW

n=12; ~1.0g body fat deficit), IWATx + Sham EWAT LIPX (10 Rev n=14, FW n=10, BW n=12; ~ 2.0g body fat deficit), EWATx + IWATx (10 Rev n=12, FW n=12, BW n=10; ~3.0g body fat deficit), Sham EWATx + Sham IWATx (10 Rev n=12, FW n=12, BW n=10; no body fat deficit). *Surgery*

All surgeries were performed under isofluorane anesthesia. EWATx was accomplished by making abdominal incision through which both EWAT pads could be accessed. The fat pads were removed, with care taken to preserve the blood vessels supplying the testes. IWATx was accomplished by making cutaneous incisions made over the lateral thigh through with both IWAT pads could be accessed and removed. The Sham surgeries consisted of exposing, but not removing, the fat pads.

Following recovery of surgery the behavioral measures began. Body mass was measured weekly and leptin was measured ever four weeks. At the end of the 14 weeks, at a time when we previously showed that LIPX-induced compensation in fat pad mass of the non-excised WAT is complete (Mauer and Bartness, 1997), the animals were decapitated, fat pads were removed (inguinal, epididymal, retroperitoneal and subcutaneous) and fat pad mass was recorded.

Data were analyzed as percent change compared with the sham LIPX group within each foraging group. The behavioral data was analyzed by analysis of variance for repeated measures with a Lipectomy x Group design for each behavioral measure. The Number Cruncher Statistical Systems (NCSS) 2001 and Pass Trial computer software, developed in Kaysville, Utah, was used to analyze the data. Fat pad mass terminal measures were analyzed by analysis of variance with a Lipectomy x Group design. Using the same stat package, comparisons between post hoc groups were made using Duncan's New Multiple-Range tests as appropriate. Planned comparisons of carcass lipid and tissue masses were made between groups with t-tests. Results were considered statistically significant if p<0.05.

Results

Foraging effort and Compensation

The animals that were required to forage for their food (10 Revolutions/pellet) did not increase IWAT mass after EWATx (Figure 1). As indicated by the significantly decreased total fat pad masses, compensation for lipid deficit was not complete in any of the foraging groups (Figure 1, 2 and 3). The graded lipid deficit was maintained at the end of the study – that is the more fat that was removed initially, the larger the remaining lipid deficit.



* p<0.05 different among all foraging groups; + p<0.05 different among all lipectomy conditions

Figure 1. 10 Revolution group animals, within each LIPX, individual fat pads are being compared to the sham animals group within the 10 Revolution foraging group



* p<0.05 different among all foraging groups; + p<0.05 different among all lipectomy conditions

Figure 2. FW group animals, within each LIPX, individual fat pads are being compared to the sham animals within the FW group



* p<0.05 different among all foraging groups; + p<0.05 different among all lipectomy conditions

Figure 3. BW group animals, within each LIPX, individual fat pads are being compared to the sham animals within the BW group.

Foraging, Food Hoarding and Food Intake

A larger lipid deficit did not trigger proportionally greater increases in foraging, food hoarding or food intake (Figure 4, 5 and 6). Increasing the foraging effort differentially affected food hoarding and food intake within the three foraging groups, specifically with the 10 Revolutions/pellet and BW groups increasing hoarding more than food intake (Figure 5 and 6). The FW group increased food intake more than food hoarding (Figure 5 and 6).



Wheel Revolutions

* p<0.05 different among all foraging groups; ## p<0.05 different from 10Revs/pellet and BW

Figure 4. Wheel revolutions were compared as percent change in each foraging group and LIPX group to the sham group within each group.



*p<0.05 different among all foraging groups; ## p<0.05 different from 10Revs/pellet and BW

Figure 5. Food hoarding was compared as percent change in each foraging group and LIPX group to the sham group within each group.



*p<0.05 different among all foraging groups; ## p<0.05 different from 10Revs/pellet and BW

Figure 6. Food intake was compared as percent change in each foraging group and LIPX group to the sham group within each group.

Discussion

The results of the present experiment showed that there was an affect of foraging effort on appetitive and consummatory ingestive behavior, there was no overall and specific fat pad compensation and there was no effect of LIPX on appetitive or consummatory ingestive behavior.

When foraging effort was increased, food hoarding was increased more so than food intake (Day and Bartness, 2001). In the present study, I found that the foraging effort also affected both appetitive and consummatory ingestive behaviors. Thus, when the hamsters were required to forage for food (10 Revolutions/pellet), food hoarding was increased to a greater extent than food intake in LIPX animals. The present study replicated the results of Day and Bartness's experiment. Furthermore, when animals were required to forage for their food (10 Revolutions/pellet), the normal compensatory increase in IWAT mass, after EWAT LIPX, (Mauer and Bartness, 1997) was inhibited.

The graded lipectomy-induced lipid deficit was still apparent in the total fat pad mass after 14 weeks, a result that differs from the Mauer and Bartness study (Mauer and Bartness, 1997) where the level of fat pad mass compensation was positively correlated to the lipectomyinduced lipid deficit. The foraging effort may be a reason why the difference occurs. The requirement of running on the wheel for pellets may have triggered the overall decrease in body mass.

The magnitude of the lipid deficit did not correspond to a proportional change in appetitive or consummatory ingestive behaviors. It was shown in a previous study that LIPX increases food hoarding in Siberian hamsters (Wood and Bartness, 1997). Although the magnitude of the lipid deficit did not trigger proportional changes in foraging, food hoarding or food intake within the present study; perhaps loss of lipid in individual fat pads may be more important in regulating appetitive and consummatory behaviors. It may be that the fat pads that are associated with critical functions such as reproduction are monitored rather than total body fat.

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