The Effects of Menstrual Cycle Phases and Adiposity on Energy Balance in Women

A closer look at variations in female sex-steroid hormones, leptin levels, the occurrence and severity of premenstrual syndrome and food reinforcement across the menstrual cycle

Jessica McNeil

Master's Thesis

University of Ottawa

Faculty of Health Sciences

Department of Human Kinetics

© Jessica McNeil, Ottawa, Canada, 2011

ABSTRACT

Energy intake (EI) and energy expenditure (EE) across the menstrual cycle (MC), while considering body adiposity, have not been previously evaluated in the same individuals. This study mainly examined the variations in energy balance (EB) across MC. Seventeen women (Body fat-DXA:28.5%) participated in three identical sessions during distinct phases of the MC: Early-follicular, Late-follicular/ovulation and Mid-luteal (confirmed by basal temperature and sex-steroid hormones). EI, resting metabolic rate (RMR), physical-activity EE (PAEE), severity of PMS, leptin and relative-reinforcing value (RRV) of preferred foods were measured during each phase. No differences in body fat, EI, RMR, PAEE, leptin and RRV of food were noted across MC. Trends were noted in preferred snack ($p=0.06$) and combined snack/fruit ($p=0.06$) intakes, while differences were noted in severity of PMS ($p<0.05$) across phases. Changes in EB across the MC were not noted. PMS was more severe, and preferred snack and combined snack/fruit intakes were slightly higher during mid-luteal phase.

Keywords: Menstrual cycle, energy intake, resting metabolic rate, physical activity energy expenditure, premenstrual syndrome, relative reinforcing value of food, leptin

TABLE OF CONTENTS

ABSTRACT 2

CHAPTER 1 – INTRODUCTION 6

1.1 ENERGY BALANCE 6

1.2 THE OCCURRENCE AND SEVERITY OF PMS, FOOD REINFORCEMENT AND LEPTIN LEVELS 7

1.3 MENSTRUAL CYCLE AND ADIPOSITY 8

1.4 DEFINITIONS 10

1.5 LIST OF ABBREVIATIONS 12

1.6 LIMITATIONS, DELIMITATIONS AND ASSUMPTIONS 13

CHAPTER 2 – LITERATURE REVIEW 14

2.1 VARIATIONS IN FEMALE SEX-STEROID HORMONES 14

2.2 VARIATIONS IN ENERGY AND MACRONUTRIENT INTAKES 16

2.3 POSSIBLE FACTORS FOR ALTERED ENERGY AND MACRONUTRIENT INTAKES 21

2.4 ENERGY EXPENDITURE 30

2.5 POSSIBLE MECHANISMS FOR ALTERED ENERGY BALANCE IN OVERWEIGHT AND OBESE WOMEN 32

2.6 CONCLUDING COMMENTS 34

2.7 OBJECTIVE 35

2.8 PRIMARY HYPOTHESES 36

2.9 SECONDARY HYPOTHESES 36

CHAPTER 3 - METHODOLOGY 37

3.1 PARTICIPANTS 37

3.2 DESIGN AND PROCEDURE 38

3.3 MEASUREMENTS 42

3.4 STATISTICAL ANALYSES 48

CHAPTER 4 - RESULTS 50

4.1 CHARACTERISTICS OF THE PARTICIPANTS 50

4.2 HORMONE LEVELS AND BASAL TEMPERATURE 52

4.3 ENERGY AND MACRONUTRIENT INTAKES 56

4.5 THE OCCURRENCE AND SEVERITY OF PMS 64

4.6 THREE FACTOR EATING QUESTIONNAIRE 66

4.7 SENSITVITY TO REWARD AND THE RELATIVE REINFORCING VALUE OF FOOD 66

CHAPTER 5 - DISCUSSION 71

5.1 ASSESSING PROPER TESTING TIMES: MEASURES OF BASAL TEMPERATURE AND SEX-STEROID HORMONES 72

5.2 ANTHROPOMETRY 73

5.3 ENERGY INTAKE AND ENERGY EXPENDITURE 74

5.4 SECONDARY FACTORS: LEPTIN LEVELS, THE OCCURRENCE AND SEVERITY OF PMS AND THE RELATIVE REINFORCING VALUE OF PREFERRED FOODS 78

CHAPTER 6 - LIMITATIONS AND FUTURE PERSPECTIVES 86

CHAPTER 7 - CONCLUSION 91

REFERENCES **92**

APPENDIX 1: **108**

PARTICIPANT CONSENT FORM **108**

Supervisor's signature 114
Principal investigator's Signature 114

APPENDIX 2: **115**

FOOD MENU AND MACRONUTRIENT BREAKDOWN **115**

APPENDIX 3: **118**

SENSITIVITY TO REWARD QUESTIONNAIRE **118**

APPENDIX 4: **122**

THREE-FACTOR EATING QUESTIONNAIRE **122**

PART 2 125

APPENDIX 5: **129**

SHORTENED PREMENSTRUAL ASSESSMENT FORM **129**

CHAPTER 1 – INTRODUCTION

1.1 ENERGY BALANCE

The menstrual cycle is a repeated, natural condition seen in women of reproductive age, which is characterized by fluctuations in female sex-steroid hormones, variations in body temperature, as well as the presence of menstrual bleeding (Johnson, Corrigan, Lemmon, Bergeron, & Crusco, 1994; Davidsen, Vistisen, & Astrup, 2007). Numerous studies have noted an effect of variations in female reproductive hormones throughout the menstrual cycle on different aspects of the energy balance (Johnson et al., 1994; Davidsen et al., 2007; Dalvit, 1981; Buffenstein, Poppitt, McDevitt, & Prentice, 1995; Dye & Blundell, 1997). These hormonal variations are not only responsible for controlling the menstrual cycle, but they may also influence energy intake, expenditure and storage in order to prepare the body in the event of a possible pregnancy (Davidsen et al., 2007). In previous animal and human studies, it has been observed that energy intake (EI) decreases during the late follicular and ovulation phases of the menstrual cycle, which are characterized by higher levels of estrogen, while EI tends to increase during the luteal phase, following ovulation, at which time plasma levels of both estrogen and progesterone are elevated (Johnson et al., 1994; Davidsen et al., 2007; Dalvit, 1981; Buffenstein et al., 1995; Dye & Blundell, 1997; Li, Tsang, & Lui, 1999; Czaja, 1978; Dalvit-McPhillips, 1983; Gong, Garrel, & Calloway, 1989; Manocha, Choudhuri, & Tandon, 1986; Martini, Lampe, Slavin, & Kurzer, 1994; Tarasuk & Beaton, 1991; Lissner, Stevens, Levitsky, Rasmussen, & Strupp, 1988; Lyons, Truswell, Mira, Vizzard, & Abraham, 1989). Variations in energy expenditure (EE) throughout the menstrual cycle have also been observed in past studies, which have

indicated increases in total EE during the luteal phase (Davidsen et al., 2007; Buffenstein et al., 1995; Dye & Blundell, 1997; Solomon, Kurzer, & Calloway, 1982; Webb, 1986; Israel & Schneller, 1950).

1.2 THE OCCURRENCE AND SEVERITY OF PMS, FOOD REINFORCEMENT AND LEPTIN LEVELS

There are certain secondary factors that may in part explain these variations in EI and EE seen across the different phases of the menstrual cycle. Among those are the occurrence and severity of the premenstrual syndrome (PMS), which occurs approximately one week prior to menstruation, and has been shown to increase EI and appetite during this period (Davidsen, 2007; Buffenstein, 1995; Dye & Blundell, 1997). In addition, another important factor to consider, which may partly explain the increases in cravings and sweet-fatty food intake during the luteal phase, is related to the possible changes in reward driven behaviors across the menstrual cycle. Interestingly enough, certain studies have shown increases in reward signaling and responsiveness in certain reward centers of the brain during the late follicular and ovulation phases of the menstrual cycle (Van Vugt, 2009; Frank, Kim, Krzemien, & Van Vugt, 2010). Finally, significant variations in leptin levels, a hormone which is secreted by adipocytes and circulates in the plasma at concentrations relative to fat mass, have been noted across the menstrual cycle (Hardie, Trayhurn, Abramovich, & Fowler, 1997; Riad-Gabriel, Jinagouda, Sharma, Boyadjian, & Saad, 1998; Al-Harithy, Al-Doghaither, & Abualnaja, 2006). Additionally, normally menstruating women have higher levels of leptin in comparison to both men and postmenopausal women even after adjustments for body fat content (Mannucci et al., 1998; Rosenbaum et al., 1996). It has also

been suggested that leptin may play an important role in the maintenance of proper reproductive functions in premenopausal women, by acting as a mediator between the levels of body fat and the onset of menstruation every month (Dye & Blundell, 1997).

1.3 MENSTRUAL CYCLE AND ADIPOSITY

Most of the studies evaluating appetite and energy variations across the menstrual cycle have focused on women with a body mass index (BMI) within the normal, recommended range (18.5-24.9 kg/m²). No studies to date have evaluated the variations in energy balance across the menstrual cycle in overweight and obese women, while only one study (Al-Harithy et al., 2006) compared female reproductive hormones and leptin levels in both lean and obese women. In addition, no studies to date have taken into consideration the possible effects of different adiposity levels, instead of BMI, on variations seen in energy balance across the different phases of the menstrual cycle, as well as measure EI, macronutrient intakes and different aspects of EE in the same individuals across the menstrual cycle. Point in fact; the variations in energy balance and female sex-steroid hormone levels in women with high adiposity levels may differ from those seen in lean women. It may be hypothesized that women with high adiposity levels may be more prone to more frequent episodes of overeating and a higher overall EI during the luteal phase, which may then possibly lead to a gradual cyclic weight gain in this population. In addition, the occurrence of PMS and the changes in reward driven behaviors or the relative reinforcing value (RRV) of food across the menstrual cycle may not only be in part responsible for a higher EI during the luteal phase, but may also contribute to the occurrence of more frequent episodes of overeating during this phase in women with high adiposity levels. Finally, given

the important role of leptin in regulating EI, EE and energy storage, as well as its substantial variations related to adiposity levels, it was thus important to evaluate EI and EE across the menstrual cycle in women with a body fat percentage above 32% (overweight/obese), in comparison to women with a body fat percentage below 32% (lean).

1.4 DEFINITIONS

1) Adiposity level: The total relative (fat percentage) or absolute (fat mass) quantity of fat that is found in one's adipose tissues.

2) Premenstrual syndrome (PMS): This syndrome often occurs during the late luteal phase or approximately 1 week prior to menses. This syndrome is classically characterized by the appearance of slight depression, restlessness, mood swings, irritability and stress, breast tenderness, bloating, as well as changes in appetite and food intake.

3) Relative reinforcing value of food: Describes how hard an individual is willing to work in order to obtain a specific food item by measuring responses at a predetermined reinforcement schedule. In this case, the specific food items are the participant's favorite snack and favorite vegetable or fruit. This task is *relative-reinforcing* since there are alternatives to choose from and the results or responses to one item will affect the results or responses to the other item.

4) Non-homeostatic factors: These include different cognitive, hedonic and environmental factors, which may influence our choices in food consumption. For instance, our choices in food consumption may be influenced by location and time of day.

5) Homeostatic signaling: Hormones which have been coined short- and long-term feeding signals, and which regulate energy intake through their interactions with neurons of the central nervous system, namely in the hypothalamus.

6) Reward driven behaviors: A psychological process that is contingent on a reinforcing stimulus. These are goal oriented choices made by the higher cortical association areas of the brain, and are based on our past experiences and positive associations that we make with specific types of foods. For instance, if eating a specific type of food provides a good, pleasurable experience, then the behavior related to consuming this food will be reinforced and later repeated.

7) Early Follicular phase: In the present study, this phase of the menstrual cycle represents the first 5 days of the cycle, with day 1 being the first day of menses. The Follicular phase represents the first half of the menstrual cycle, or days 1-13 based on a 28-day cycle.

8) Late follicular/Ovulation: In the present study, this phase of the menstrual cycle represents the last 3 days of the follicular phase and 1 day of ovulation, based on a 28-day cycle. Ovulation usually occurs at mid-point during the menstrual cycle, or day 14 based on a 28 day cycle.

9) Mid-Luteal phase: In the present study, this phase represents the time at which progesterone levels are elevated, which included days 21-26 based on a 28-day cycle. The Luteal phase represents the second half of the menstrual cycle, or days 15-28 based on a 28-day cycle.

1.5 LIST OF ABBREVIATIONS

BMI: Body mass index

CIA: Chemiluminescence immunoassay

CMIA: Chemiluminescent microparticle immunoassay

DXA: Dual-energy X-ray absorptiometry

ECLIA: Electrochemiluminescence immunoassay

EDTA: Ethylenediaminetetraacetic acid

EE: Energy Expenditure

EI: Energy Intake

ELISA: Enzyme-linked immunosorbent assay

FSH: Follicule stimulating hormone

GnRH: Gonadotropin-releasing hormone

HA: High adiposity

kcal: Kilocalories

LH: Luteinizing hormone

n: number of participants

PAEE: Physical activity energy expenditure

PMS: Premenstrual syndrome

RMR: Resting metabolic rate

RRV: Relative reinforcing value

SD: Standard deviation

TFEQ: Three-Factor Eating Questionnaire

1.6 LIMITATIONS, DELIMITATIONS AND ASSUMPTIONS

Over the course of this study, it was assumed that the participants followed the protocol as prescribed, which included not eating for at least 12 hours prior to the start of testing, as well as no structured physical activity (i.e. playing sports and training) and alcohol consumption for at least 24 hours prior to the start of testing. The participants chose to commit to these conditions by accepting the conditions in the consent form and signing the latter. It is also assumed that the participants followed the instructions given to them while outside of the laboratory, meaning that they truthfully and properly reported values of food and beverage intakes in the food journals and wore the accelerometers for the allocated times. Finally, it was assumed that the participants truthfully answered all questionnaires given to them throughout the study. The present study was limited to a sample of lean (Body fat percentage of 29% or lower) and overweight (Body fat percentage above 29%), non-smoking, university aged women.

CHAPTER 2 – LITERATURE REVIEW

2.1 VARIATIONS IN FEMALE SEX-STEROID HORMONES

The female sex-steroid hormones that are mainly responsible for initiating and controlling the menstrual cycle are the gonadotropin-releasing hormone (GnRH), the follicule stimulating hormone (FSH), the luteinizing hormone (LH), estrogen and progesterone (Davidsen et al., 2007). Following this further, the GnRH is secreted by the hypothalamus and acts on the pituitary gland. At this point, the pituitary gland produces two types of gonadotropins, known as FSH and LH. These gonadotropins then act on the ovaries, while stimulating the production and release of reproductive hormones. The main reproductive hormones produced by the ovaries in women are estrogen and progesterone; the fluctuations in their release aiding in the determination of the different menstrual cycle phases, known as the follicular phase, ovulation, and the luteal phase (Davidsen et al., 2007; Farage, Neill, & Maclean, 2009).

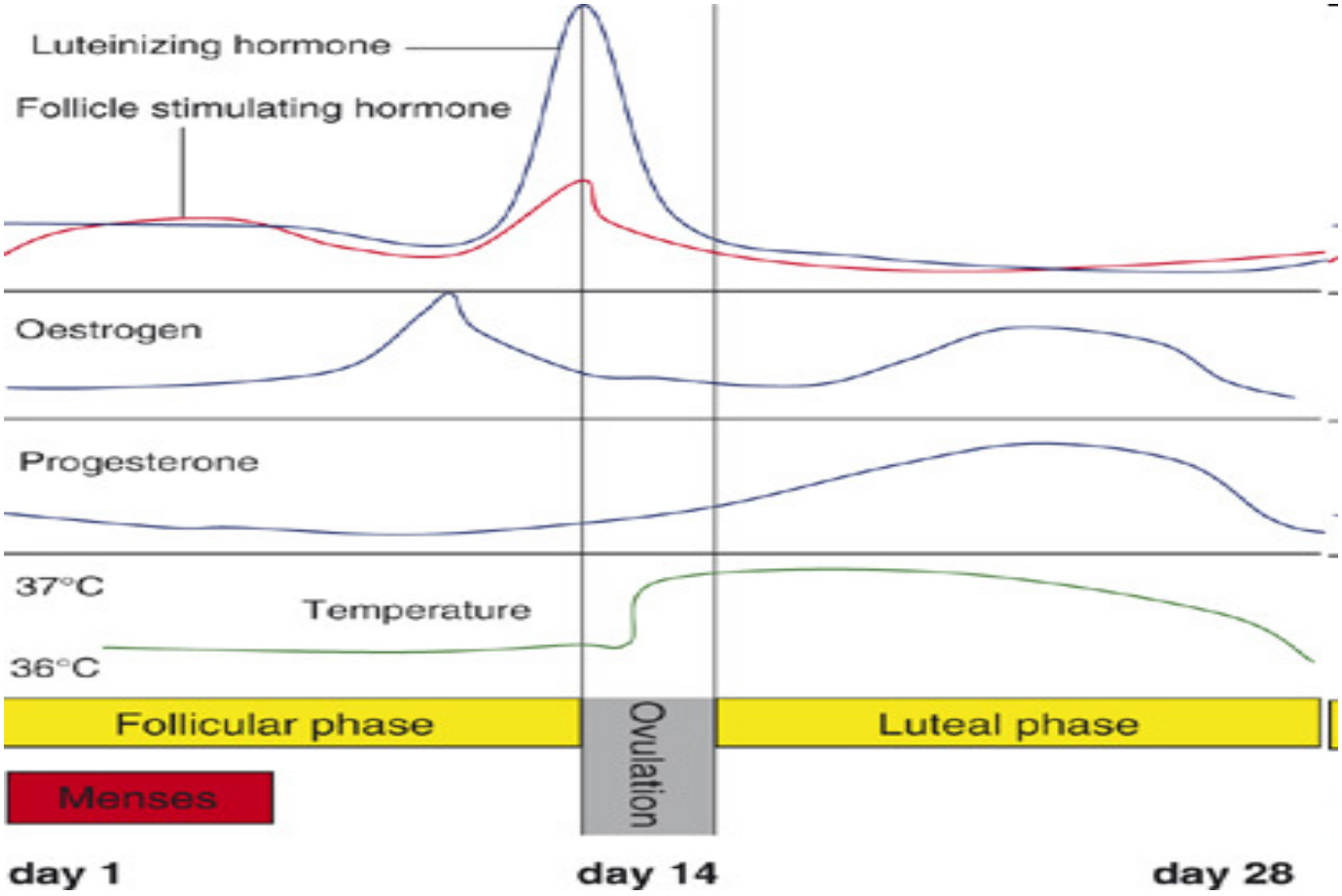


FIGURE 1. Normal changes in hormones and body temperature across the menstrual cycle.

Source: Davidsen L, Vistisen B, Astrup A. Impact of the menstrual cycle on determinants of energy balance : a putative role in weight loss attempts. International Journal of Obesity 2007;31:1777-85.

As shown in **Figure 1**, during the early follicular phase (or menses), there are low levels of plasma LH, estrogen and progesterone, but high circulating levels of FSH, which facilitates follicular growth. This surge in FSH provokes an increase in estrogen levels during the mid follicular phase; this hormone attaining a peak at the end of this phase before starting to decrease. This peak in estrogen then triggers the surge in LH during mid-cycle, lasting about 40-48 hours and producing ovulation. Following ovulation, progesterone levels increase, and reach a peak during the mid-luteal phase. This phase is also characterized by a slight increase in estrogen levels. Finally, plasma levels of estrogen and progesterone decrease at the end of the cycle, which then initiates menstruation and the beginning of a new menstrual cycle (Davidsen et al., 2007; Buffenstein et al., 1995; Farage et al., 2009).

2.2 VARIATIONS IN ENERGY AND MACRONUTRIENT INTAKES

The fluctuations in female sex-steroid hormones observed across the menstrual cycle seem to induce an overall variation in EI through changes in hunger levels, cravings for certain foods, adjustments in meal sizes, the occurrence of snacking, as well as variations in macronutrient intake (Johnson et al., 1994; Buffenstein et al., 1995; Dye & Blundell, 1997). More specifically, many studies have observed a decrease in EI during the follicular phase, in relation to the increase in estrogen levels, as well as an increase in EI during the luteal phase, which is characterized by an increase in progesterone levels. A fairly large variation can be observed when comparing the caloric intake values reported by different studies, presented in **Table 1**, which evaluated the variations in EI across the menstrual cycle; with

increases in EI ranging from 87-500 kcal during the luteal phase in comparison to the follicular phase.

Table 1. Variations in Energy Intake across the menstrual cycle in lean women.

				Energy Intake (kcal)				
Study	**Days measured**	**Food assessment method**	***N***	**M**	**F**	**O**	**L**	**Δ between F and L**
Li et al., 1999	Days 6-10 after onset of menses and days 6-10 after ovulation	Self-monitored food records	20	-	1455.53	-	1666.67	211.14
Johnson et al., 1994	One complete menstrual cycle	Self-monitored food records	26	1815.04	1709.52		1873.36	163.84
Dalvit et al., 1981	Two complete menstrual cycles	Self-monitored food records	8	-	1439.88	-	1939.81	499.93
Lissner et al., 1988	Days 1-10 and 19-28	Ad libitum selection and weighed food	23	-	2248	-	2335	87
Lyons et al., 1989	35 days	Self-monitored food records	18	2155	2012	1874	2198	186
Dalvit-McPhillips, 1983	10 days pre- and 10 days post-ovulation	Dietary recall	8	-	1246.78	-	1724.47	477.69
Gong et al., 1989	One complete menstrual cycle	Self-monitored food records	7	1884.49	1831.95	1765.07	2039.74	207.79
Manocha et al., 1986	10 days pre- and 10 days post-ovulation over 2	Self-monitored food records	11	-	1299.32	-	1609.82	310.50

	complete menstrual cycles							
Martini et al., 1994	3 days pre- and 3 days post-ovulation	Self-monitored food records	18	-	1748.35	-	1905.99	157.64
Tarasuk and Beaton, 1991	10 days pre- and 10 days post-ovulation	Self-monitored food records	14	-	1820.01	-	1910.77	90.76

Note: *N*, Number of participants; M, Menses; F, Follicular phase; O, Ovulation; L, Luteal phase; Δ, difference.

Certain studies have also specified that EI is at its lowest point during ovulation, following the peak in plasma estrogen levels (Davidsen et al., 2007; Dalvit, 1981, Buffenstein et al., 1995; Dye & Blundell, 1997; Czaja, 1978; Lyons et al., 1989). However, even though clear variations in EI have been previously observed in lean women (**Table 1**), it is important to note that no significant changes in body weight or fat percentage during the different phases of the menstrual cycle have been noted in this population (Johnson et al., 1994; Davidsen et al., 2007; Buffenstein et al., 1995; Dye & Blundell, 1997; Li et al., 1999; Riad-Gabriel et al., 1998). Taking this into consideration, it has been shown that EI changes in relation to estrogen levels, with this hormone having a clear inhibitory effect on appetite and food consumption (Davidsen et al., 2007; Dye & Blundell, 1997; Czaja, 1978; Lyons et al., 1989). Czaja and Goy (1975) have specifically demonstrated this by observing a significant depression in food intake in ovariectomized primates who received estradiol injections, which created a surge in estrogen levels similar to those seen during the menstrual cycle. Czaja (1978) also demonstrated that peak estradiol levels occurred between days 15 to 18 before menses, while minimum food intake occurred between days 15 to 19 before

menses in intact cycling female rhesus monkeys. Following this further, it is relevant to note that the female rhesus monkey's menstrual cycle is similar to human menstrual cycles in both duration and hormone levels, permitting to compare the data obtained in monkeys to humans (Evans & Foltin, 2004). Even though the relationship between estrogen levels and EI seems apparent, the impact of progesterone levels on food intake is not as clear. Certain studies have demonstrated that progesterone is not an appetite stimulant and does not directly affect food intake. According to Czaja (1978) and Hess and Resko (1973), progesterone in intact, regularly cycling female primates seems to have a secondary effect on food intake, by decreasing and inhibiting the effects of circulating estrogen. Thus, the attenuation in the effects of circulating estrogen levels on food intake, through the inhibitory actions of progesterone, could then explain the prevention of a decrease in food intake during the luteal phase due to estrogen. However, these variations in female reproductive hormones do not explain the increase in EI observed during the luteal phase. Point in fact; even though clear variations in EI have been previously observed in lean women (**Table 1**), no significant changes in body weight or fat percentage during the different phases of the menstrual cycle have been noted in this population (Johnson et al., 1994; Davidsen et al., 2007; Buffenstein et al., 1995; Dye & Blundell, 1997; Li et al., 1999; Riad-Gabriel et al., 1998), possibly meaning that the increase in EI seen during the luteal phase may be compensated by the decrease in EI seen during the follicular phase (Davidsen et al., 2007; Buffenstein et al., 1995). However, there may be different or even more pronounced variations in EI in women with high adiposity levels, considering that this population may be more vulnerable to more frequent episodes of overeating, as well as having a relatively

higher EI during the luteal phase of the menstrual cycle. This may ultimately affect cyclic body weight and/or fat percentage in these individuals.

In addition to variations in total EI, certain studies have also noted changes in macronutrient intake across each distinct phase. These variations are shown in **Table 2**.

Table 2. Variations in Macronutrient Intake across the menstrual cycle in lean women.

	Macronutrient Intake (% Energy Intake)											
	Carbohydrate				**Fat**				**Protein**			
Study	**M**	**F**	**O**	**L**	**M**	**F**	**O**	**L**	**M**	**F**	**O**	**L**
Li et al., 1999	-	51	-	52	-	29	-	31	-	19	-	18
Johnson et al., 1994	47.9	47.5		47.9	35.8	35.4		37.4	15.1	15.4		14.7
Lyons et al., 1989	47	48	48	47	36	35	36	36	13	14	14	14
Dalvit-McPhillips, 1983	-	39.9	-	55.8	-	44.4	-	32.6	-	15.7	-	11.6
Martini et al., 1994	-	51.3	-	50.1	-	33.1	-	34.2	-	15.6	-	15.6
Terasuk and Beaton, 1991	-	43.8	-	43	-	36	-	37.6	-	14.6	-	14.1

Note: M, Menses; F, Follicular phase; O, Ovulation; L, Luteal phase.

Most of these studies noted similar results, in which case increases in relative carbohydrate and fat intakes are seen prior to menses, while relative protein intake seems to decrease during this time. It is also not surprising to note that these alterations in macronutrient intake seem to be directly related to changes in total EI seen across the

menstrual cycle given the high density of fat and sugar. In the present study, energy and macronutrient intakes were directly measured inside the laboratory over the course of 3 days (1 day/menstrual cycle phase) with food menus, as well as outside the laboratory with 3-day food journals (1 journal/menstrual cycle phase).

2.3 POSSIBLE FACTORS FOR ALTERED ENERGY AND MACRONUTRIENT INTAKES

2.3.1 The occurrences and severity of the premenstrual syndrome (PMS)

An important factor which seems to affect a great number of women is the occurrence of premenstrual syndrome (PMS) in the late luteal phase. This syndrome is classically characterized by the appearance of slight depression, restlessness, mood swings, irritability and stress, breast tenderness, bloating, as well as changes in appetite and EI (Farage, Osborn, & MacLean, 2008; Bryant, Truesdale, & Dye, 2006). Approximately 50% of women suffer from a minimal level of distress related to PMS, from which 30.6%, 13.6% and 8.1% of women reported low, moderate and severe levels of distress respectively (Angst, Sellaro, Merikangas, & Endicott, 2001). Based on these findings, it seems that certain symptoms which characterize PMS affect a great number of women, but to various degrees (Dye & Blundell, 1997; Johnson, Carr-Nangle, & Bergeron, 1995; Dye, Warner, & Bancroft, 1995; Cross, Marley, Miles, & Wilson, 2001; Both-Orthman, Rubinow, Hoban, Malley, & Grover, 1988). More specifically, women who reported more severe PMS symptoms consumed on average more calories, and presented more frequent episodes of overeating and cravings for sweet-fatty foods during the late luteal phase (Davidsen et al., 2007; Dye & Blundell, 1997; Dye et al., 1995; Both-Orthman et al., 1988; Masho, Adera, & South-Paul, 2005; Michener, Rozin, Freeman, & Gale, 1999; Rogers & Smit, 2000). In addition, one study (Cross et al., 2001) showed that women who suffered from PMS had

significantly higher EI for each macronutrient during the luteal phase compared to the follicular phase, while women without PMS only showed significant differences in fat intake between these two phases. After adjusting for EI, the women who suffered from PMS also showed significant increases in fat, carbohydrate and simple sugar intake, while the women without PMS showed no significant differences between macronutrient intakes during the follicular and luteal phases. There were also no significant differences between the groups in relation to age, BMI and menstrual cycle length. Wurtman, Brzezinski, Wurtman, & Laferrere (1989) have also noted interesting findings, where the consumption of a high-carbohydrate, low-protein meal during the late luteal phase alleviated depression, anger, tension, confusion, sadness, fatigue, and improved alertness and calmness in women who reported suffering from PMS. No effects of this meal consumption were observed in these women during the follicular phase, as well as in women who did not report suffering from PMS during either phase.

2.3.2 Reward driven behaviors and the relative reinforcing value of food

Another interesting factor which may in part explain the increases in sweet-fatty food cravings during the luteal phase is related to the changes in reward driven behaviors across the menstrual cycle. Generally speaking, reward driven behaviors are goal oriented choices driven by the higher cortical association areas of the brain, and are based on our past experiences and positive associations that we make with specific types of foods (Rogers & Smit, 2000; Cameron & Doucet, 2007). And so, if eating a specific type of food provides a good, pleasurable experience, then the behavior related to consuming this food will be reinforced and later repeated. In addition, these chosen eating behaviors are often influenced

by non-homeostatic factors, as opposed to homeostatic signaling, where different cognitive, hedonic and environmental factors may greatly influence our choices in food consumption (Cameron & Doucet, 2007).

In relation to the menstrual cycle, the increase in overall EI, especially sweet-fatty food intake, during the luteal phase may be in part related to a more pronounced impact of certain non-homeostatic factors on one's eating behavior choices. In a study done by Pliner & Fleming (1983), pleasure sensations were measured before and after a glucose load in 34 women during the mid-follicular and mid-luteal phases. The absence of hunger sensation, related to a decrease in the reported pleasantness rating following a glucose load, was noted during the mid-luteal phase only. Taking these results into consideration, the presence of hunger does not seem to explain the increase in total EI previously discussed during the luteal phase (**Table 1**). However, based on these findings, it may be hypothesized that homeostatic signals related to food consumption and hunger may influence women's eating behaviors during the follicular phase of the menstrual cycle, while the non-homeostatic factors, or food reward, related to certain foods may play a larger role in deciding the quantity and quality of foods consumed during the luteal phase of the menstrual cycle. Increases in overall EI and cravings for sweet-fatty foods may also be related to decreases in reward signaling initiated by the consumption of these types of foods during the luteal phase. These observations are supported by a study (Van Vugt, 2009) which investigated the activation of the corticolimbic structures, or reward centers of the brain, to visual food cues during each phase of the menstrual cycle. Through the use of functional MRI scans, they observed a more elicit response to both high and low calorie visual food cues in most of the

reward centers of the brain during the late follicular and ovulation phases. Based on these observations, an increase in reward signaling during the late follicular and ovulation phases may actually lead to a decrease in EI since the reward centers of the brain may be more sensitive to feelings of satisfaction following the consumption of a specific food. On the other hand, a decrease in reward signaling during the luteal phase may lead to a more pronounced EI in order to obtain this same level of satisfaction. In addition to the previous findings, the participants of a related study (Frank et al., 2010) rated high calorie foods as being more appealing than low calorie foods during menstruation and the luteal phase (weeks 1, 3 and 4), but not during the late follicular phase (week 2). And so, the increase in preferences for low calorie foods during the late follicular phase may also contribute to a lower EI during this phase.

There seems to also be a relation between the occurrence of PMS and reward driven behaviors. Freeman, Schweizer and Rickels (1995) used the Tridimensional Personality Questionnaire (TPQ), which measures harm avoidance, novelty seeking and reward dependence, in women who suffer from moderate to severe levels of PMS. They observed a slight, but not significant, increase in reward dependence scores. They also noted a significant increase in harm avoidance and novelty seeking scores in women with PMS, with high novelty seeking personality traits correlating with premenstrual food cravings, headaches, mood swings and anxiety (Freeman et al., 1995). It is known that self-reporting appetite and cravings for different high dense, palatable foods have been associated with negative moods, such as tension, anger, depression and tiredness (Rogers & Smit 2000; Wurtman & Wurtman, 1989), which are certain symptoms that characterize PMS. However,

it has also been shown that even if women report having cravings for chocolate prior to menses, they do not necessarily eat more chocolate at this time (Jas, 1996).

Finally, certain anorexigenic hormones such as leptin and insulin have been coined long-term feeding signals; both of which are related to adiposity level and, consequently, regulate EI through their interactions with certain neurons of the central nervous system (Cameron & Doucet, 2007; Figlewicz & Benoit, 2009). More specifically to leptin, it is predicted that under-nutrition and over-nutrition would respectively increase and decrease the rewarding value of food, in order to maintain proper body weight and adiposity level (Zheng & Berthoud, 2007). This hormone also seems to affect the rewarding value of food by modulating the sensitivity of taste receptor cells in the oral cavity (Shigemura et al., 2004), alter olfactory detection (Julliard et al., 2007) and influence the visual perception of food (Uher, Treasure, Heining, Brammer, & Campbell, 2006). However, even though relative increases in leptin have an anorexigenic effect, leptin signaling is not able to prevent overconsumption of food short-term, simply because the differences in EI from one meal to the next is usually very small when taking whole body energy reserves into consideration (Mela & Rogers, 1998). In addition, many non-homeostatic factors, such as different cognitive, hedonic and environmental factors, may override the long-term metabolic feeding signals (Cameron & Doucet, 2007; Zheng & Berthoud, 2007), and may then ultimately affect one's choices in food consumption.

In summary, the rewarding aspect of foods does take on a great importance in the current environment where many types of food are readily available. As such, the

suppression of short-term homeostatic signaling may lead to an overconsumption of certain foods; with this factor seeming to play a predominant role during the luteal phase of the menstrual cycle.

2.3.3 Leptin levels

Certain studies have shown that leptin, a hormone which is mostly secreted by white adipose tissue and varies in response to changes in energy storage (Murphy & Bloom, 2004), may also play an important role in initiating and regulating reproductive events (Hardie et al., 1997; Riad-Gabriel et al., 1998; Al-Harithy et al., 2006; Mannucci et al., 1998; Rosenbaum et al., 1996; Cunningham, Clifton, & Steiner, 1999; Thong, McLean, & Graham, 2000; Chehab, Lim, & Lu, 1996; Chehab, Mounzih, Lu, & Lim, 1997). Higher levels of leptin have been observed in women of reproductive age, in comparison to men and postmenopausal women after adjusting for body fat content (Rosenbaum et al., 1996). Moreover, in a study performed on obese sterile female mice (Chehab et al., 1996), the administration of human recombinant leptin injections not only corrected obesity in these female mice, but also restored full fertility in the latter by stimulating the production of LH in the pituitary gland and increasing the number of primary and mature vesicular follicles. In a similar study (Chehab et al., 1997), the administration of exogenous leptin in prepubertal female mice of normal weight has shown to accelerate the onset of puberty in the latter. In women, a sufficient amount of fasting leptin concentration, which is based on energy availability, is required for the onset of puberty and the maintenance of proper reproductive functions (Cunningham et al., 1999; Thong et al., 2000; Licinio et al., 1998; Kopp et al., 1997; Goumenou, Matalliotakis, Koumantakis, & Panidis, 2003). More specifically, one

study (Welt et al., 2004) indicated that a critical serum leptin level of 1.2 and 1.85 ng/ml is required to initiate the release of FSH and LH respectively, meaning that serum leptin levels lower than these may lead to the occurrence of primary or secondary amenorrhea.

It is also possible to assume that leptin levels may also vary in relation to the different phases of the menstrual cycle. It has been demonstrated that fasting leptin levels are at their lowest point during the late luteal to mid-follicular phases inclusively, to then begin increasing during the late follicular phase, and eventually peak during the early to mid-luteal phase, with values approximately 35-60% higher at this time, in comparison to those observed in the early follicular phase (**Table 3**). On the other hand, certain studies have noted no significant variations in leptin levels across the menstrual cycle (Capobianco et al., 2010; Mills, Ziegler, & Morrison, 1998; Teirmaa, Luukkaa, Rouru, Koulu, & Huuppomen, 1998).

Table 3. Variations in leptin levels across the menstrual cycle.

				Leptin levels (ng/ml)			
Study	**Days measured**	**Assessment of leptin levels**	***N***	**M levels or day 3**	**F levels; % variation compared to M**	**Post-O levels; % variation compared to M**	**Mid-L levels; % variation compared to M or F**
Mannucci et al. 1998	Days 3, 10, 17 and 24	Fasting blood sample	18	11.2	13.8; ↑ 23%	14.67; ↑31%	15.12; ↑35% (M)
Riad-Gabriel et al. 1998	Every other day from days 1-9 and days 17-28; Every day from days 10-16.	Fasting blood sample	9	14.9	-	-	20.4; ↑51%
Hardie et al. 1997	Day 2, and every 3rd day afterwards; Every day from days 11-17.	Blood sample	6	22.9	27.5; ↑20%	-	36.7; ↑ 60% (M)
Al-Harithy et al. 2006	Days 3, 10, 17 and 24	Fasting blood sample	65 Obese:	10.6	7.7; ↓38.3	9.60; ↓25.5	12.67; ↑19.75 (M)
			Lean:	6.70	8.0; ↑20.5	7.77; ↑15.97	10.01; ↑49.40 (M)
Thong et al. 2000	One pre- and one post-ovulation	Fasting blood sample	21 Elite athlete:	-	3.0	-	4.38; ↑46% (F)
			Recreat-ionally active:	-	6.0	-	8.4; ↑40% (F)
Mills et al. 1998	One pre- and post-ovulation	Blood sample	30	-	15.4	-	17.1; ↑10% (F)
Capobianco et al. 2010	One menses, one pre-ovulatory and one luteal	Blood sample	18	10	13; ↑23% (M)	-	11; ↑9% (M)
Teirmaa et al. 1998	One menses, one ovulatory and one luteal	Fasting blood sample	8	10.2	10.7; ↑5% (M)	-	11.8; ↑14% (M)

Note: *N*, Number of participants; M, Menses; F, Follicular phase; Post-O, Post-Ovulation; Mid-L, Mid-Luteal phase.

Interestingly enough, the increase in leptin levels seen during the late follicular phase to early luteal phase is not related to changes in body fat in lean women. The exact reasons

behind these variations in leptin levels across the menstrual cycle are not yet well known. Nevertheless, certain studies have hypothesized possible reasons which may in part influence the surge in leptin levels seen during the luteal phase. First and foremost, it has been hypothesized that the variations in leptin levels may be possibly mediated by the changes in female sex-steroid hormones, especially those of estrogen. It has been noted that the variations in leptin levels in women throughout life follow a similar pathway as that of estrogen, significantly rising at the onset of puberty and then decreasing after menopause (Hardie et al., 1997), suggesting that there might be a close relationship between leptin and estrogen. Estrogen receptors have also been found in white adipose tissue (Goumenou et al., 2003), suggesting that this hormone may play a direct role in leptin secretion. Additionally, one study (Shimizu et al., 1997) noted that leptin levels were decreased by ovariectomy in rats, while estradiol supplement later reversed the effects of ovariectomy on these circulating leptin levels, thus leading to an increase in leptin levels. Finally, the changes in leptin levels across the menstrual cycle have been correlated with that of estrogen levels (Riad-Gabriel et al., 1998), with the late follicular phase peak in estrogen levels being possibly responsible for the stimulation of leptin secretion observed during the early luteal phase (Hardie et al., 1997; Al-Harithy et al., 2006; Mannucci et al., 1998). With regard to the possible effects of progesterone on leptin secretion, these two hormones show similar patterns of fluctuations across the menstrual cycle, with both hormones attaining high levels during the mid-luteal phase (Al-Harithy et al., 2006), and certain studies have also noted a strong correlation between progesterone and leptin values during the luteal phase (Hardie et al., 1997; Al-Harithy et al., 2006; Goumenou et al., 2003). However, only correlations have been observed between leptin and progesterone values, without identifying any possible cause-effect

relationship. In summary, it seems more likely that high estrogen levels may be in part responsible for the increase in leptin secretion seen across the menstrual cycle. However, the exact effects of either estrogen or progesterone levels on leptin secretion are still unclear and seem to be very complex according to present literature (Hardie et al., 1997; Riad-Gabriel et al., 1998; Al-Harithy et al., 2006; Mannucci et al., 1998; Thong et al., 2000; Goumenou et al., 2003; Groschl et al., 2002; Kitawaki et al., 2000).

A second hypothesis postulates that higher leptin concentrations in the luteal phase may play a significant role in preparing the body for the metabolic demands of pregnancy (Riad-Gabriel et al., 1998; Goumenou et al., 2003; Cioffi et al., 1997). It has been previously demonstrated that leptin levels significantly increase during pregnancy, to a point where a state of leptin resistance occurs in order to maintain an elevated level in EI through the prevention of the satiety effect of this hormone, while encouraging the accumulation of fat storage during the 2nd and 3rd trimesters (Augustine, Ladyman, & Grattan, 2008; Ladyman, 2008). This being said, a more subtle version of this leptin resistance state may take place during the luteal phase, in order to favor an increase in EI and prepare energy reserves for the possible occurrence of pregnancy (Cunningham et al., 1999). Finally, it has also been suggested that the rise in leptin levels during the mid-luteal phase may in fact simply reflect, rather than affect, the changes in caloric intake (Van Vugt, 2009).

2.4 ENERGY EXPENDITURE

The possible variations in resting metabolic rate (RMR) and physical activity EE (PAEE) across the menstrual cycle have been previously addressed. First, variations in

RMR, sleeping metabolic rate and 24-hour EE across the menstrual cycle have been noted, with increases mainly occurring after ovulation and during the mid-luteal phase (Davidsen et al., 2007; Buffenstein et al., 1995; Solomon et al., 1982; Webb, 1986). The mean increase in 24-hour EE following ovulation, in comparison to the follicular phase, is about 2.5-11.5%, or approximately 89-279 kilocalories (Davidsen et al., 2007; Webb, 1986). However, most studies have also noted inter-individual variations of EE across the menstrual cycle, in which case not all participants demonstrated an increase in 24-hour EE following ovulation (Davidsen et al., 2007; Solomon et al., 1986; Webb, 1986). Hence, many factors, which may or may not be related to the reproductive system, seem to contribute to this variation observed in EE. Moreover, even though 24-hour EE may increase by about 89-279 kilocalories during the luteal phase in comparison to the follicular phase (Davidsen et al., 2007; Webb, 1986), variations in EI during this phase have been noted to be similar or even higher (**Table 1**). This increase in EE may then, in some cases, be insufficient to account for the increase seen in EI (Solomon et al., 1982), thus possibly creating a positive energy balance during this phase of the menstrual cycle. As for the practice of structured and non-structured physical activity, no variation in this aspect of EE has been noted across the different phases of the menstrual cycle through the use of physical activity journals (Johnson et al., 1994). However, the practice of voluntary exercise has been proven to attenuate the severity of certain PMS symptoms, and was negatively correlated with water retention, episodes of dizziness and nausea, as well as increases in appetite (Johnson et al., 1995). The present study evaluated certain aspects of EE, by specifically measuring and analyzing the changes in RMR, as well as PAEE through the use of accelerometers across the different phases of the menstrual cycle.

2.5 POSSIBLE MECHANISMS FOR ALTERED ENERGY BALANCE IN OVERWEIGHT AND OBESE WOMEN

Granted the documented variations in EI and EE across the menstrual cycle, as well as the possible causes that underlie them, it is important for the purpose of this thesis to consider these variations in relation to body adiposity. In previous studies, no significant changes in body weight or fat percentage across the menstrual cycle have been noted in women with a BMI within the normal, recommended range (Johnson et al., 1994; Davidsen et al., 2007; Dalvit, 1981; Buffenstein et al., 1995; Dye & Blundell, 1997; Li et al., 1999; Lissner et al., 1988; Lyons et al., 1989). However, to date, not much is known regarding the effects of menstrual cycle fluctuations on energy balance in overweight and obese women. In addition, it is not known whether women with high adiposity levels display different variations in EI and EE, which may ultimately lead to cyclic changes in body weight and/or fat percentage in this population. Even though this topic has not been well investigated yet in this population, certain underlying factors related to the menstrual cycle, and which may alter eating behavior, may affect overweight and obese women differently in comparison to their lean counterparts.

2.5.1 The prevalence and severity of PMS symptoms

One of the important underlying factors is related to the prevalence and severity of PMS symptoms, which may lead to episodes of overeating and, ultimately, weight gain over time. As mentioned previously, severe PMS sufferers are more prone to increasing their EI during the late luteal phase. This factor may be especially important to consider in

overweight and obese women. Indeed, Masho et al. (2005) have noted an increase in the prevalence of PMS in relation to increasing BMI, identifying obese women as being nearly three times more likely to suffer from PMS symptoms compared to normal weight women. Another study (Hartz, Barboriak, Wong, Katayama, & Rimm, 1979) also noted that women who are heavier have a higher prevalence of having irregular cycles lasting longer than 36 days, abnormal ovulations, as well as heavier menstrual flows. It has also been noted that overweight and obese individuals are more likely to be sleep deprived, stressed, depressed and exercise less, all factors which may lead to more frequent episodes of snacking and overeating (Johnson et al., 1995).

2.5.2 The reinforcing value of food

A second underlying factor which may be responsible for an increase in snacking and overall EI in overweight and obese women is related to the reinforcing value of food. As described earlier, the reinforcing value of food can be generally described as the amount of work and effort one individual is willing to do in order to obtain a certain type of food (Lappalainen & Epstein, 1990). In a study by Saelens & Epstein (1996), it was demonstrated that eating snack foods was more reinforcing for obese women in comparison to non-obese women, and these same individuals were also willing to work harder in order to obtain a snack food versus engaging in a sedentary activity. Moreover, the obese women consumed more calories when given the snack food than did the non-obese women. And so, not only do obese individuals find food more reinforcing, in which case they work harder for food than for sedentary activities (Saelens & Epstein, 1996), but they also find high-fat foods to be more reinforcing than low-fat foods when compared to lean individuals (Epstein,

Bulik, Perkins, Caggiula, & Rodefer, 1991). To our knowledge, no study to date has evaluated the changes in the reinforcing value of food in lean and overweight/obese women during the different phases of the menstrual cycle, which is why the present study aimed to evaluate the reinforcing value of each participant's favorite snack food vs. favorite fruit or vegetable during each phase of the menstrual cycle.

2.6 CONCLUDING COMMENTS

There are many studies which have attempted to evaluate the different aspects which constitute energy balance variations across the menstrual cycle, as well as the many causes which may underlie these variations. In many instances, EI and EE appear to be higher during the luteal phase, in comparison to the follicular phase. In addition, the increases in food cravings or, more specifically, sweet-fatty food intake during the luteal phase may be in part due to the occurrence and severity of PMS, as well as the changes in reward driven behaviors which may take place at this time. While taking this into consideration, the vast majority of the reported studies have only focused on women with a BMI within the normal recommended range. To our knowledge, no study to date has evaluated the variations in EI and EE across the menstrual cycle in overweight and obese women, which emphasizes the matter that it is important to pursue future studies on this particular subject in this population. It may be hypothesized that women with high adiposity levels may be more vulnerable to more frequent episodes of overeating, as well as have an overall higher EI during the luteal phase, in comparison to women with an adiposity level within the recommended range, which may possibly lead to a gradual cyclic weight gain in this

population. It has been previously demonstrated by many studies that no significant variations in body weight in lean women occur across the menstrual cycle (Johnson et al., 1994; Davidsen et al., 2007; Dalvit, 1981; Buffenstein et al., 1995; Dye & Blundell, 1997; Li et al., 1999; Lissner et al., 1988; Lyons et al., 1989), meaning that the increase in EI seen during the luteal phase is most likely compensated for by the decrease in EI observed during the follicular phase (Davidsen et al., 2007; Buffenstein et al., 1995). However, if there is a significantly higher EI in women with high adiposity levels during the luteal phase, in comparison to lean women, this resulting positive energy balance may not be able to be accounted for during the follicular phase, therefore leading to a gradual cyclic weight gain in this population. In addition to this, the findings of this study may have a good practical significance, by providing useful information when building an adequate weight management program for premenopausal women, which not only takes into account certain physiological mechanisms related to feeding and the menstrual cycle, but which also tailors to an overweight/obese population.

2.7 OBJECTIVE

The primary objective of this study was to evaluate the variations in EI, macronutrient intake and EE in women with different adiposity levels. The secondary objective of this study was to evaluate the effects and variations in female sex-steroid hormones, leptin, the occurrence and severity of PMS and preferred food reinforcement.

2.8 PRIMARY HYPOTHESES

1) Women with high adiposity levels will have the greatest variation in EI across the menstrual cycle.

2) There will be no significant differences in the relative values of RMR between women with different adiposity levels across the menstrual cycle.

3) Women with high adiposity levels will significantly decrease their EE from physical activities during the luteal phase of the menstrual cycle.

2.9 SECONDARY HYPOTHESES

1) Both groups will present similar variations in patterns and values in all female sex-steroid hormone levels across their cycle. Leptin levels in both groups will show a similar variation in pattern, while being significantly higher during the luteal phase. However, in women with high adiposity levels, this hormone will be significantly higher during each phase of the cycle.

2) Women with high adiposity levels will have a higher score on the shortened PMS assessment form during the luteal phase.

3) Women with high adiposity levels will show stronger food reinforcement for snack foods, especially during the luteal phase of the menstrual cycle.

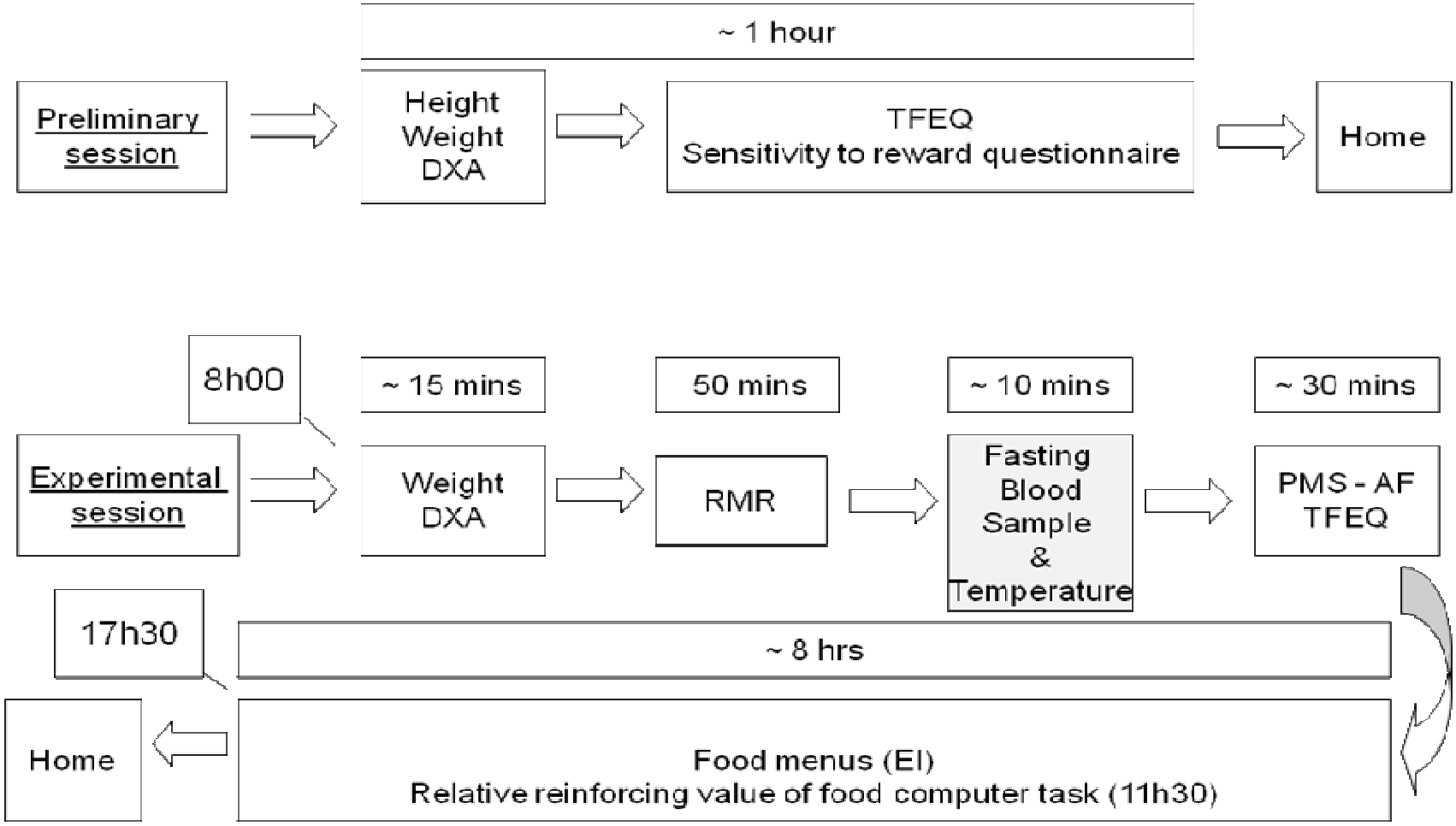
CHAPTER 3 - METHODOLOGY

3.1 PARTICIPANTS

A total of 18 women participated in this study. However, one participant had to be excluded due to her female sex-steroid hormone levels being well below the normal range (Bermant & Davidson, 1974), suggesting that an anovulatory cycle had occurred in her case. And so, the results of 17 participants are presented herein. The recruited participants had to adhere to certain selection criterions: 1) between the ages of 18-40 years; 2) Has had a stable weight (± 2 kg) within the past 6 months; 3) Has not used any form of medical contraceptives (pill, patch, injection) within the past 6 months; 4) Was not currently taking any antibiotics or prescribed medication. The exclusion criterions for this study were: 1) the use of any form of medical contraceptives; 2) professional athletes or women in the midst of training for a competition; 3) women who had a BMI below 18.5 kg/m^2; 4) smokers; 5) women who suffered from type 2 diabetes; 6) women who had menstrual cycle irregularities within the past 6 months (changes in menstrual cycle lengths or absence of menses). The body fat percentage results obtained during each experimental session were used to classify the women according to body adiposity. In this sample, the cut-off point, based on a median split, was 29% body fat, in which case nine participants had 29% or less body fat (lean or normal adiposity level) and eight participants had more than 29% body fat (high adiposity level). This study was conducted according to the guidelines laid down in the Declaration of Helsinki and all the procedures involving human participants were approved by the University of Ottawa ethics committee. Written informed consent was also obtained from all participants.

3.2 DESIGN AND PROCEDURE

This study followed a within-subjects repeated measures design. An outline of the study's design and procedures is presented in **Figure 3**. All tests were performed at the Behavioral and Metabolic Research Unit (BMRU) in Ottawa between October 2010 and April 2011. Baseline measurements were taken during the preliminary session in order to determine whether each participant corresponded to the inclusion criterions and to obtain pre-experimental measurements of each participant. Additionally, three identical experimental sessions were conducted, with each experimental session being held during a specific phase of the menstrual cycle (early follicular, late follicular/ovulation and mid-luteal). Each participant was asked to count the number of days (length) of her menstrual cycle for at least 1 month prior to testing; this permitted us to tailor the times of testing for each participant. And so, the testing sessions were scheduled according to the length of each participant's menstrual cycle. Plasma sex-steroid hormone levels and basal temperature measured during each experimental session also aided in confirming each menstrual cycle phase.



TFEQ: Three-factor eating questionnaire
RMR: Resting metabolic rate
PMS – AF: shortened PMS assessment form

FIGURE 2. Outline of the study design and procedures.

3.2.1 Preliminary session

During this time, the participants were informed of the experimental procedures which were employed during each experimental session, as well as the equipment which were used in order to obtain the necessary data. Following this information session, a consent form was completed and signed by the participants. Afterwards, the weight and height of the participants were measured. In addition, body composition was measured using DXA. The participants were also required to complete the Three-Factor Eating Questionnaire (TFEQ) (Stunkard & Messick, 1985) and the sensitivity to reward questionnaire (Franken & Muris, 2005). This session lasted approximately one hour.

3.2.2 Experimental sessions: Overview of the timeline

Following the baseline measurements, a total of three identical laboratory sessions for each participant were held over one complete menstrual cycle, in order to be able to test the stated hypotheses. More specifically, each laboratory session lasted approximately 10 hours, and was held once during the early follicular phase (days 1-5 inclusively of the cycle), once prior or during the time of ovulation (days 10-14 inclusively), as well as once during the mid-luteal phase (days 22-26 inclusively). It is important to note that the days of the menstrual cycle associated with each phase are based on a 28-day cycle and were adjusted according to the length of each participant's menstrual cycle. In addition, not all participants had their first session during the early follicular phase; this was the case for four participants, while eight participants had their first session during the late follicular/ovulation phase and six started with the mid-luteal phase testing session.

3.2.3 Experimental sessions: Anthropometric and temperature measurements, resting metabolic rate, blood samples, relative reinforcement value of food task and questionnaires

The participants arrived at the laboratory at 8h00 following a 12-hour overnight fast, and restrained from any form of structured physical activity (e.i. training or playing sports) and alcohol consumption for at least 24 hours prior to the beginning of each experimental session. At this moment, body weight and body composition were measured. Afterwards, the participants laid and rested for 20 minutes (without falling asleep) before testing RMR using indirect calorimetry (Vmax encore 29N, Viasys respiratory care Incorporated, Palm Springs, California, USA). The RMR test itself lasted 30 minutes. After this test, a fasting blood sample was taken in order to evaluate the plasma levels of estrogen, progesterone, FSH, LH and leptin during this phase of the menstrual cycle. Basal temperature was also measured orally with a digital thermometer. Afterwards, a shortened premenstrual assessment form (Allen, McBride, & Pirie, 1991) was answered by the participants in order to determine the severity of PMS symptoms at this time (during this phase of their menstrual cycle). The participants also completed the TFEQ (Stunkard & Messick, 1985) in order to determine her level of dietary restraint, dishinibition and perceived hunger during this phase of her menstrual cycle. At 11h30, the participants completed the RRV of food computer task (Lappalainen & Epstein, 1990).

3.2.4 Experimental sessions: Measurements of energy intake and physical activity energy expenditure

The participants stayed in a room inside the laboratory for the day (9h30-17h30) and consumed all their meals and snacks in this room, and were also allowed to do any type of

sedentary activities that they desired during the day. Once settled in this room, the participants were handed a copy of the food menu, which included a wide variety of foods and beverages, from which they chose the type of food from the menu that they may want to consume (McNeil, Riou, Razmjou, Cadieux, & Doucet, 2011). At this time, the chosen foods were prepared and served to the participants in a sufficient amount (2 portions of each item). This food menu was first presented to the participants at 9h30, then at 10h30, 12h30, 14h30, 15h30 and finally 16h30. At the end of each experimental session, which was at 17h30, a 3-day food journal and an accelerometer were given to the participants in order to record food intake and PAEE outside of the laboratory setting. Each participant was instructed on how to properly use each tool, and she brought back the food journal and the accelerometer at her next experimental session. The participants then left the laboratory at approximately 18h00.

3.3 MEASUREMENTS

3.3.1 Sensitivity to reward questionnaire (Preliminary session only)

All participants were asked to complete the 35-item sensitivity to reward questionnaire (Torrubia, Avila, Molto, & Caseras, 2001), which measured each individual's views on the rewarding value of different factors (e.i. money, food, social connections) and how these factors impact their decisions, which may in turn affect a person's EI and eating behavior. The test-retest reliability results obtained on the sensitivity to reward questionnaire at baseline, 3 months, 1 year and 3 years were respectively 0.87, 0.69 and 0.61 (Torrubia et al., 2001). See **Appendix 4** for a copy of the sensitivity to reward questionnaire.

3.3.2 Anthropometric and temperature measurements

Standing height was measured to the nearest centimeter using a wall stadiometer, Tanita HR-100 height rod, without shoes (Tanita Corporation of America, Inc, Arlington Heights, IL). Body weight and body composition were measured using a standard beam scale (HR-100; BWB-800AS, Tanita Corporation, Arlington Heights, IL., USA) and DXA scanner (Lunar Prodigy, General Electric, Madison, WI, USA) respectively during the preliminary session, as well as during each experimental session. The coefficient of variation and correlation for body fat percentage measured by the DXA scanner in 12 healthy participants were 1.8% and $r = 0.99$ respectively. The basal temperature of the participants was measured orally through the use of a digital thermometer (rapid digital thermometer, BD, Franklin Lakes, NJ, USA). These measurements were taken at the beginning of each experimental session in order to determine whether changes in body weight, body composition and basal temperature incurred due to menstrual cycle phase.

3.3.3 Energy intake and Pleasantness rating of foods

The overall EI and macronutrient composition of foods consumed by the participants during each laboratory session were evaluated through the use of a food menu (McNeil et al., 2011). See **Appendix 3** for a copy of the food menu and the macronutrient breakdown of each item. This food menu contains 62 items, which include breakfast items, snacks, hot meals, caloric beverages and water. During each experimental session, the participants were handed a copy of the food menu on six different occasions throughout the day, at which time they were able to choose the type of foods and beverages from the menu that they may want to consume. The chosen foods and beverages were then prepared and served to the

participants. The use of a food menu inside the laboratory setting is a good way to monitor and measure the total amount of food that a person eats, while letting the latter choose the type of foods and beverages that they want to consume throughout the day. The foods chosen by the participants were weighed in grams before serving, using an electronic scale (Scout Pro SP2001, Ohaus Corporation, Pine Brook, N.J.), as well as after they were done eating, in order to precisely evaluate the amount of food that they consumed. The macronutrient composition of foods consumed were determined and analyzed through the use of nutritional labels, as well as the Food Processor SQL software (version 9.6.2; ESHA Research, Salem, OR). At the end of each experimental session, a 3-day food journal (Bingham et al., 1995) was given to the participants in order to record food intake outside of the laboratory setting. As for the measurement of food and beverage pleasantness, participants were asked to draw a vertical line on a 150 mm visual analogue scale reflecting their appreciation for all foods and beverages that they consumed during each experimental session inside the lab. The question asked on the visual analogue scale was: "How pleasant is the taste of this food?"

3.3.4 Energy expenditure

The RMR of the participants were evaluated using indirect calorimetry (Vmax encore 29N, Viasys respiratory care Incorporated, Palm Springs, California, USA) at the beginning of each experimental session, while the participants were fasting. The coefficient of variation and correlation for the RMR measured by the Vmax encore 29N system in 12 healthy participants were 5.1% and $r = 0.94$ respectively. As for PAEE, the latter was predicted through the use of an accelerometer (Actical Accelerometer, Bio-Lynx Scientific Equipment,

Montreal, Quebec, Canada) over the time span of 7 days following each experimental session. The Actical was worn on the right hip only and the logging time was set at 15 seconds. In each epoch, when the average activity count for successive 10 or 3 minutes is less than 50 counts/min, it is labeled as Sedentary (0.007565 kcal/min/kg). All activity counts above this, but lower than the first cut-off point (light-moderate), are classified as light activity. Finally, the cut-off points for the classification of each activity are 0.031 kcal/min/kg and 0.083 kcal/min/kg for light-moderate and moderate to vigorous activities respectively. A single regression model is used to calculate PAEE values; with the following equation used for activity counts equal or higher than the first cut-off point (activity EE = 0.02778 + (1.143E-5) x AC), where AC corresponds to activity count. The coefficient of variation for adults for this equation is 0.71.

3.3.5 Blood sample

A single blood sample was drawn from the antecubital vein of the non-dominant arm when the participants were fasting in order to determine the plasma levels of estrogen, progesterone, FSH, LH and leptin during each distinct phase of the menstrual cycle. Each blood sample was placed into a tube containing EDTA and was centrifuged at 3500 rpm at 4^0C immediately after the blood was drawn and stored at -80^0C until assayed. LH and FSH levels were assayed by means of a 2 step "sandwich" CIA using the Beckman Coulter Dxl Unicel 800 for serum or plasma (Beckman Coulter Canada Incorporated, Mississauga, Ontario, Canada). Progesterone levels were assayed by means of an ECLIA system, Elecsys 2010 disk system (Roche Diagnostics, Indianapolis, Indiana, USA). As for estrogen analyses, a CMIA procedure was employed through the use of an Architect estradiol reagent

kit (Abbott Laboratories, Abbott Park, Illinois, USA). 100 test reagent packs were used to analyze all sex-steroid hormone levels. Leptin levels were assayed by means of a dual range ELISA human leptin kit (Millipore Corporation, Billerica, Massachusetts, USA). The limit of sensitivity of this assay is 0.5 ng/ml for a 25 μL sample size. Leptin concentrations were determined as the average of duplicate determinations. The average duplicate coefficient of variation for plate 1 (early follicular and late follicular/ovulation phases) and plate 2 (mid-luteal phase) were 8.3 and 5.5 % respectively.

3.3.6 Three-Factor Eating Questionnaire

The participants completed the TFEQ during the preliminary and each experimental session; a 51-item validated questionnaire (Stunkard & Messick, 1985) that evaluates three aspects of eating behavior known as dietary restraint (flexible and rigid control of weight and eating habits, strategic dieting behavior, attitude to self regulation and avoidance of fattening foods), dishinibition (habitual, emotional and situational susceptibility to eating) and perceived hunger (internal and external drives for hunger). This questionnaire has been shown to be highly reliable in 220 men and women, with a coefficient of reliability of 0.9 for dietary restraint, 0.87 for dishinibition and 0.82 for perceived hunger (Stunkard & Messick, 1985). The application of this questionnaire throughout the study allowed us to evaluate whether changes in these eating behaviors may occur due to menstrual cycle phase. See **Appendix 5** for a copy of the TFEQ.

3.3.7 Occurrence and severity of PMS

The occurrence and severity of PMS was evaluated with the use of the shortened premenstrual assessment form (Allen et al., 1991). This questionnaire is used to classify the subjective changes in mood and physical conditions, based on a 6 point visual analogue scale (1 = no change and 6 = extreme change) seen or felt by the participants at the time of measurement using a ten category chart. These categories (or symptoms) include feelings of pain and tenderness, feeling stressed or sad, water retention, weight gain, ect. A score below 30 indicates none to mild changes in mood and physical conditions, a score of 30-40 indicates moderate changes, while scores of 40-50 and 50-60 indicate severe and extreme changes in mood and physical conditions respectively. It is important to note that this questionnaire classifies "PMS sufferers" as women who have either severe or extreme changes in mood and physical conditions. The test-retest reliability of this questionnaire was 0.8 in 55 women (Allen et al., 1991). See **Appendix 6** for a copy of the shortened premenstrual assessment form.

3.3.8 Relative reinforcing value of food

The RRV of a preferred snack food versus a preferred vegetable or fruit was measured through the use of a progressive ratio computer task (Lappalainen & Epstein, 1990) where the probability of earning food points varied across each trial, depending on the type of food. A small sample of the participants' favorite snack and favorite fruit or vegetable was presented to them prior to the test and they were asked to consume both samples. Following this, the participants earned food points by working for the food item of choice through the use of a slot machine program. The number of button presses needed to

obtain a point towards the preferred vegetable or fruit doubled with each trial, and this for a total of 5 trials. On the other hand, the number of button presses needed to obtain a point towards the preferred snack food exponentially increased with each trial and this also for 5 trials. And so, during the 5th trial, 10 button presses were needed in order to obtain 1 point towards the fruit or vegetable vs. 32 button presses for 1 point towards the snack. The participants then received a specific amount of each food item based on their point distribution during the test. For instance, if 100% of points were given to 1 item, the participants received 5 times the sample amount of that item vs. if 50% of points were given to each item, the participants then received both items but in lesser quantity. In order to determine the quantity of preferred food given, a ratio of 1 point for 1 slice/piece of the preferred was given; with 1 slice/piece being equivalent to approximately 4-5 grams. Please note that in the present study, all participants chose a favorite fruit, and so this type of preferred food will only be referred to as "favorite fruit" instead of "favorite fruit or vegetable" in the results and discussion sections.

3.4 STATISTICAL ANALYSES

Statistical analyses were performed using SPSS software (version 17.0; SPSS Inc, Chicago, IL). A two-way repeated measures analysis of variance was used to determine the main effects of menstrual cycle phase (within-subject factor) (early follicular, late follicular/ovulation and mid-luteal) and fat percentage (between subject factor) on the components of dietary intake (total amount of energy (kcal), protein (kcal), carbohydrate (kcal) and lipid (kcal) as well as the percent of energy from protein (%), carbohydrate (%) and lipid (%)) for the in-laboratory sessions and the 3-day food journals, hormone levels

(FSH, LH, estrogen, progesterone and leptin), RMR, PAEE, reported PMS, results on the TFEQ, as well as the RRV of snack and fruit and the associated *ad libitum* consumption of each of these preferred foods. Moreover, a two-way repeated measures analysis of variance was also used in order to determine if there were any differences between energy and macronutrient intakes directly measured inside the laboratory, as well as those reported (3-day food journal) according to body fat percentage. Bivariate correlations were also calculated between direct measurements and reported values of energy and macronutrient intakes, leptin levels, PAEE and results on the TFEQ in relation to body fat percentage, reported PMS, sex-steroid hormone levels, sensitivity to reward results and RRV of food points, button presses and *ad libitum* consumption of the preferred snack and fruit. Values are presented as means ± standard deviation. Effects with p-values < 0.05 were considered statistically significant.

CHAPTER 4 - RESULTS

4.1 CHARACTERISTICS OF THE PARTICIPANTS

The characteristics of the participants are shown in **Table 4**.

Table 4. Participant characteristics.

	Early Follicular				Late follicular/ovulation				Mid-Luteal						
	Lean		HA		Lean		HA		Lean		HA				
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Phase	Fat %	Phase* Fat %
Body weight (kg)	56.1	4.3	65.5	7.2	56.1	4.4	65.0	7.6	56.2	4.4	65.2	7.3	NS	0.006	NS
BMI (kg/m²)	21.6	1.4	23.2	1.6	21.7	1.4	23.0	1.7	21.6	1.6	23.1	1.6	NS	0.063	NS
Fat mass (%)	23.6	4.9	32.9	4.7	24.0	5.3	33.2	4.7	23.6	5.0	33.4	4.8	NS	0.001	NS
Fat mass (kg)	13.2	3.3	21.5	5.1	13.5	3.5	21.6	5.5	13.3	3.4	21.8	5.2	NS	0.001	NS
Fat-free mass (kg)	42.5	2.7	43.4	3.9	42.2	2.8	42.9	3.9	42.5	2.7	42.9	3.2	0.019	NS	NS

Note: Lean women with percent body fat of 29% or lower (n=9).

HA (high adiposity) women with percent body fat of 29.1% or higher (n=8).

The average age of the participants was 22.4±3.2 years. All participants were students following a postsecondary education and none of the participants had children. The average cycle length was 28.2±3.7 days for all participants and no significant difference in cycle length was noted between groups (data not shown). As expected, there were no significant differences in body weight, BMI, percent body fat and fat mass in all participants across the menstrual cycle. However, a significant difference was noted in fat-free mass across the menstrual cycle in all participants (**Figure 3**), where fat-free mass was found to be significantly higher during the early follicular phase in comparison to the late follicular/ovulation phase ($p<0.05$). As shown in **Table 4**, a significant difference in body weight, body fat percentage and fat mass was found between groups. A trend was also noted for BMI between groups. However, no significant interactions were noted between these characteristics and body fat percentage across the menstrual cycle. Finally, a significant difference in height (1.61±0.05 vs. 1.68±0.05 meters, $p<0.05$) was noted between groups, in which case the participants with higher adiposity levels were significantly taller.

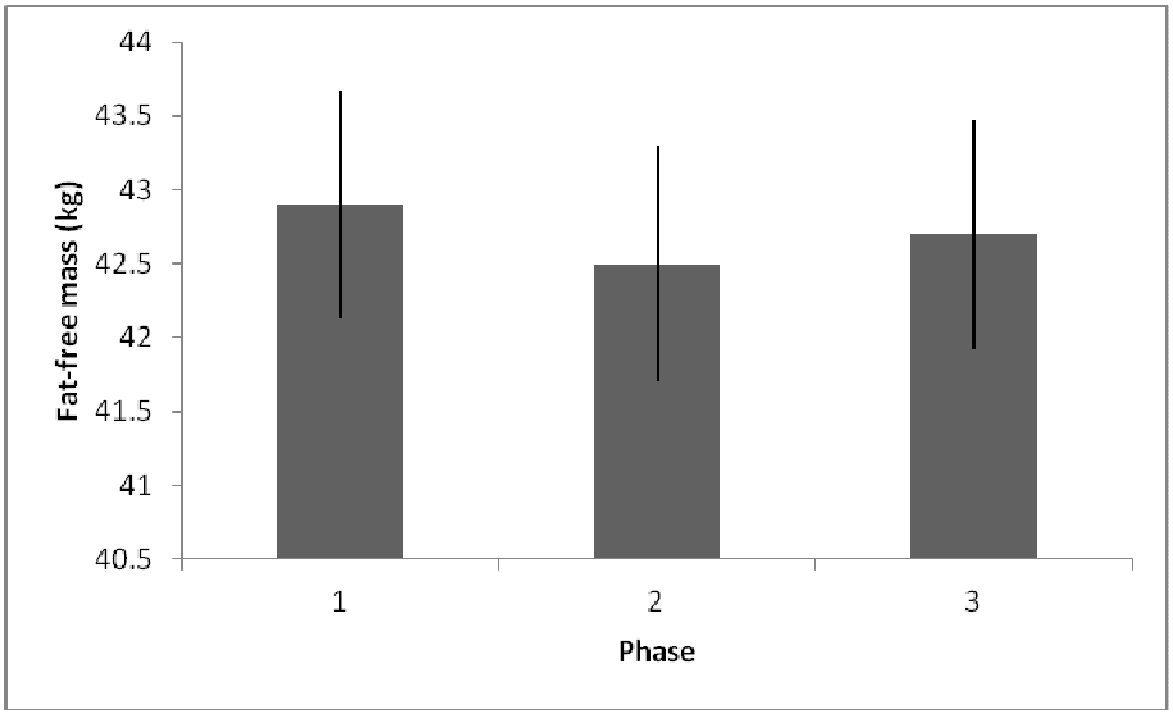
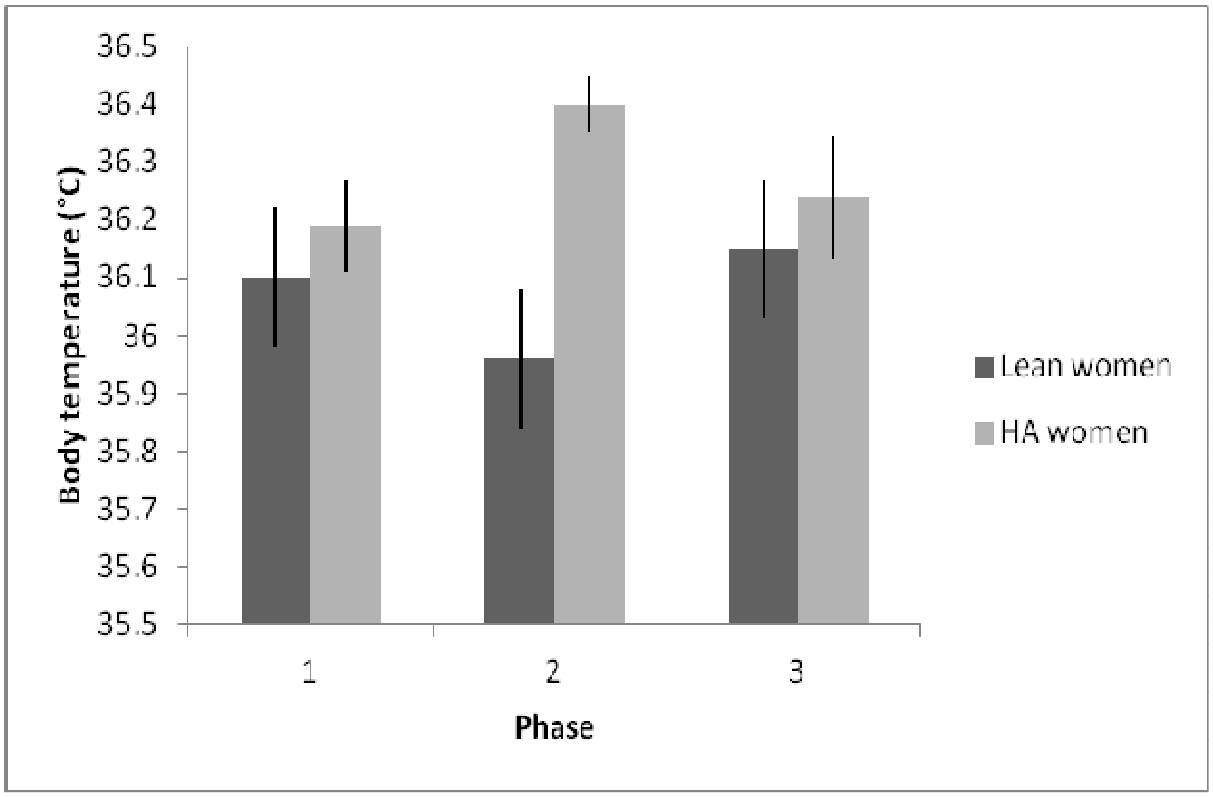


Figure 3. Variations in fat-free mass (kg) across the menstrual cycle in all participants.

Note: 1, Early Follicular; 2, Late follicular/Ovulation; 3, Mid-Luteal.

4.2 HORMONE LEVELS AND BASAL TEMPERATURE

No significant difference was noted when looking at basal temperature measured across the menstrual cycle in all participants (36.14±0.42, 36.17±0.44, 36.19±0.44°; p=NS), as well as between groups. However, a significant interaction between basal temperature and percent body fat was noted ($p<0.05$) across the cycle (**Figure 4**), where basal temperature was at its zenith during the late follicular/ovulation phase in HA women, while attaining its peak during the mid-luteal phase in lean women.



Note: 1, Early Follicular; 2, Late follicular/Ovulation; 3, Mid-Luteal.
Lean women with percent body fat of 29% or lower (n=9).
HA (high adiposity) women with percent body fat of 29.1% or higher (n=8).

Figure 4. Basal temperature measurements across the menstrual cycle according to percent body fat.

Figure 5 presents the variations in sex-steroid hormones across the menstrual cycle in all participants. Significant differences were noted for FSH (5±2, 6±2, 4±2 IU/L; $p<0.0001$), LH (4±3, 9±7, 5±5 IU/L; $p<0.05$), estrogen (93±74, 284±294, 445±154 pmol/L; $p<0.01$) and progesterone (2.7±1.0, 2.4±1.0, 37.6±25.2 nmol/L; $p<0.0001$) levels across the menstrual cycle in all participants. More specifically, significant differences were noted in FSH levels between the late follicular/ovulation and early follicular phases ($p<0.05$), as well as the mid-luteal phase ($p<0.0001$). Moreover, a trend in FSH levels was noted between the

early follicular and mid-luteal phases ($p=0.063$). As for LH levels, a significant difference was only noted between the early follicular and the late follicular/ovulation phases ($p<0.01$). Estrogen levels were significantly lower during the early follicular phase, in comparison to the late follicular/ovulation ($p<0.05$) and mid-luteal phases ($p<0.0001$). Finally, progesterone levels were significantly higher during the mid-luteal phase, in comparison to the early follicular ($p<0.0001$) and late follicular/ovulation phases ($p<0.0001$). In regard to adiposity level, no significant differences and interactions were noted for all sex-steroid hormones between groups.

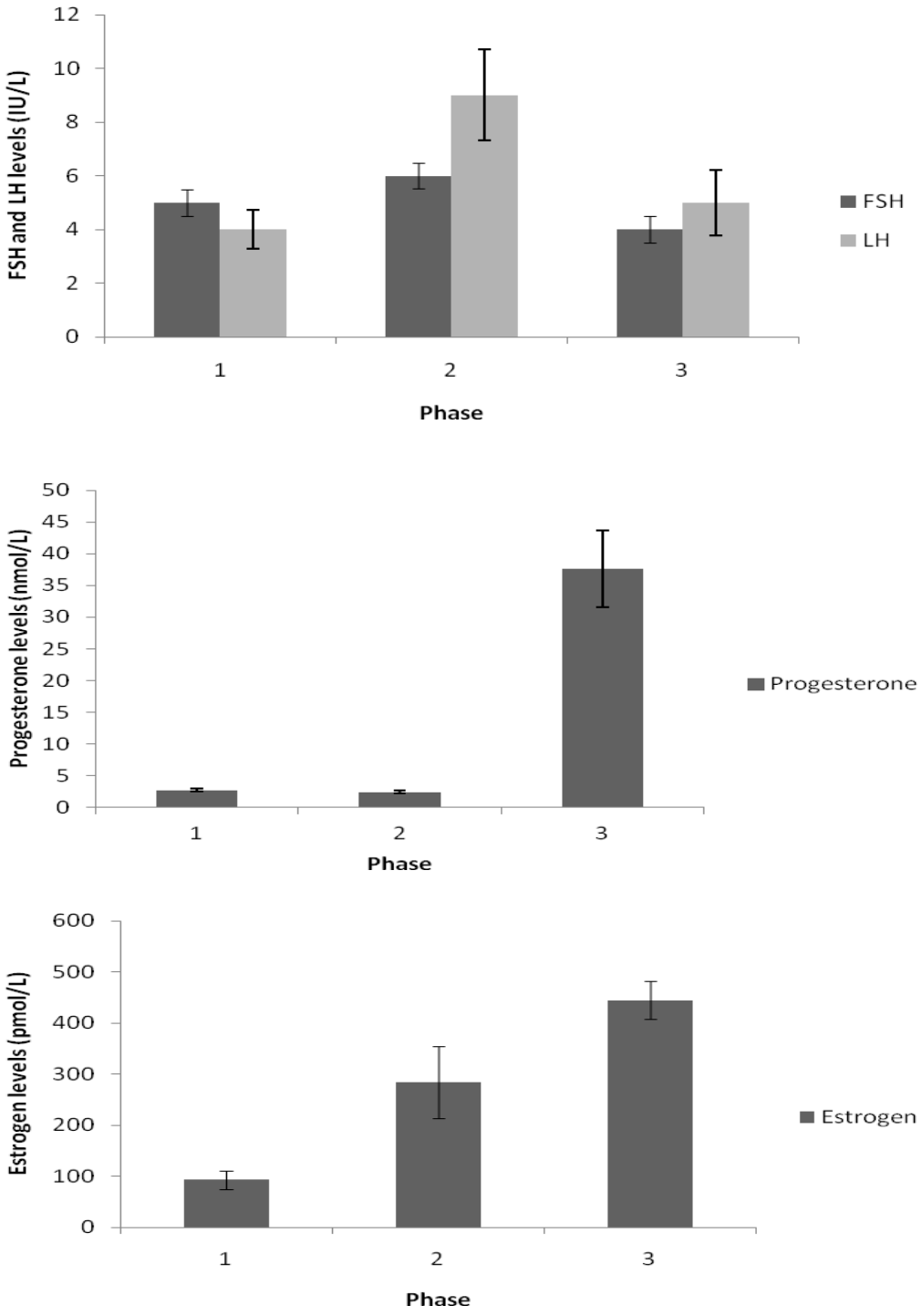


Figure 5. Variations in FSH, LH, estrogen and progesterone levels across the menstrual cycle in all participants.

Note: 1, Early Follicular; 2, Late follicular/ovulation; 3, Mid-Luteal.

No significant difference in leptin concentration (12.2±10.1, 11.3±11.1, 11.5±11.0 ng/ml; p=NS) across the menstrual cycle and interaction between adiposity, menstrual cycle phase and leptin levels were noted. However, a significant difference between groups (7.6±5.2 vs. 18.3±12.3, 5.6±2.9 vs. 19.0±13.7, 6.7±3.8 vs. 18.0±14.5 ng/ml; $p<0.05$) was noted, where the HA women had higher absolute leptin levels than did the lean women. Finally, no significant correlations were noted between leptin and sex-steroid hormone (FSH, LH, estrogen and progesterone) levels for each phase of the menstrual cycle (data not shown).

4.3 ENERGY AND MACRONUTRIENT INTAKES

As shown in **Table 5**, no significant differences were noted for total measured energy, carbohydrate, lipid and protein intakes across the menstrual cycle in lean and HA women. Moreover, no significant interactions were noted between menstrual cycle phase and body fat percentage for the in-laboratory measurements of EI. However, a significant interaction was noted between menstrual cycle phase and body fat percentage for in-laboratory protein intake only. As expected, our results also revealed that energy and macronutrient intakes in kcal tended to be higher in women with high adiposity levels; however, these differences were found to be non-significant. Finally, no significant difference was noted in pleasantness ratings of foods and beverages consumed across phases (122±17, 124±14, 120±16 mm; p=NS), suggesting that the foods and beverages consumed were equally liked and appreciated across the menstrual cycle.

Table 5. Measured energy and macronutrient intakes across the menstrual cycle (inside the laboratory).

	Early Follicular				Late follicular/ovulation				Mid-Luteal						
	Lean		HA		Lean		HA		Lean		HA				
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Phase	Fat %	Phase* Fat %
EI (kcal)	1928	475	2451	948	2101	764	2444	783	2073	527	2395	613	NS	NS	NS
Carb (kcal)	1083	181	1364	501	1149	410	1349	458	1149	346	1298	341	NS	NS	NS
Lipid (kcal)	657	264	803	416	719	355	833	317	708	205	844	305	NS	NS	NS
Protein (kcal)	236	74	316	89	286	95	293	77	266	65	292	56	NS	NS	0.049

Note: Lean women with percent body fat of 29% or lower (n=9).
HA (high adiposity) women with percent body fat of 29.1% or higher (n=8).

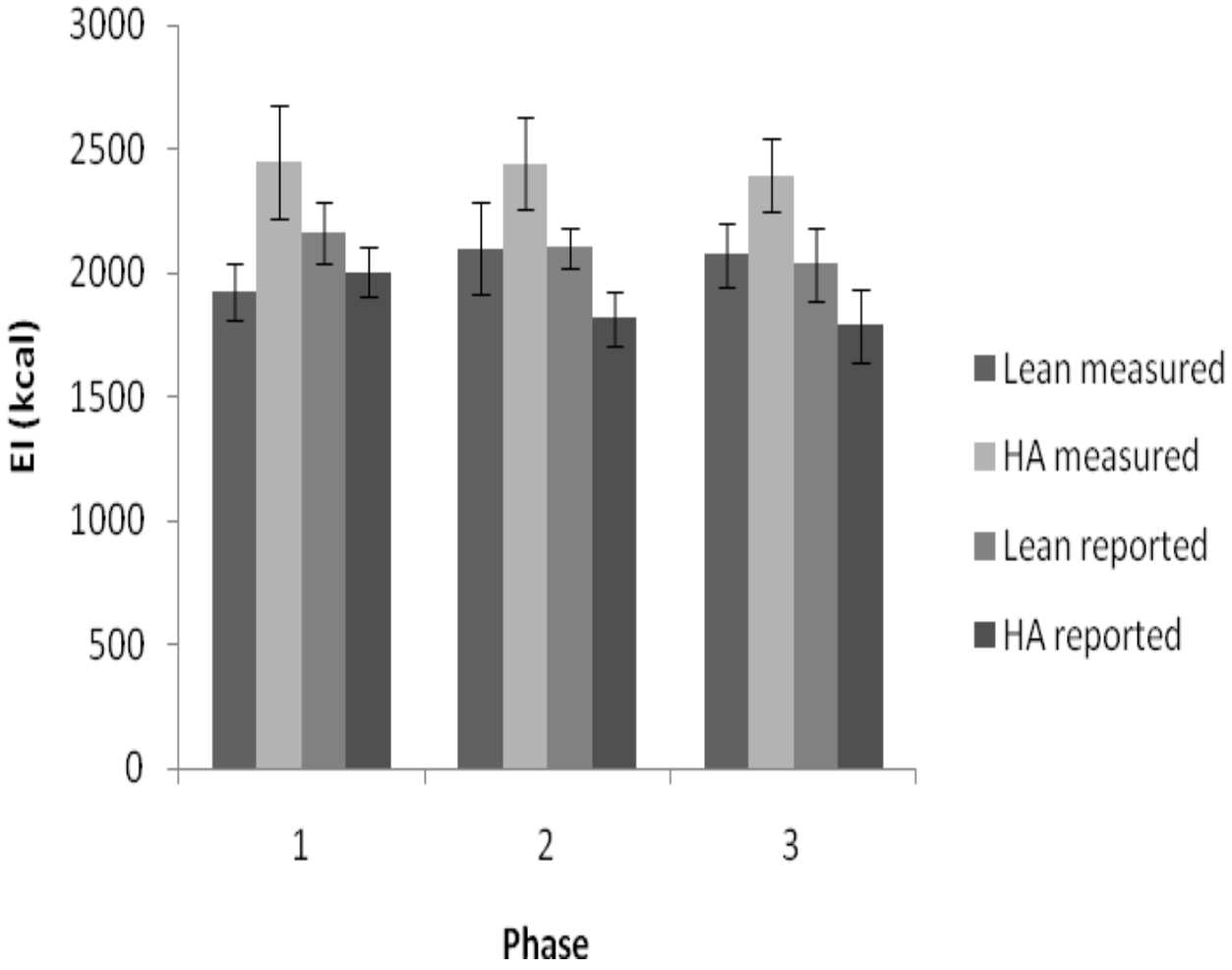
As shown in **Table 6**, no significant differences were noted for total reported energy, carbohydrate, lipid and protein intakes across the menstrual cycle in lean and HA women. Moreover, no significant interactions were noted between menstrual cycle phase and reported energy and macronutrient intakes. These results also revealed that absolute values of reported energy and macronutrient intakes, except for protein intake during the late follicular/ovulation phase, were surprisingly lower in women with higher adiposity levels, which is not in agreement with the values directly measured inside the laboratory. However, the differences in reported energy and macronutrient intakes were also found to be non-significant between groups.

Table 6. Reported energy and macronutrient intakes across the menstrual cycle (3-day food journals).

	Early Follicular				**Late follicular/ovulation**				**Mid-Luteal**						
	Lean		HA		Lean		HA		Lean		HA				
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Phase	Fat %	Phase* Fat %
EI (kcal)	2162	522	2006	407	2102	330	1819	456	2038	614	1788	604	NS	NS	NS
Carb (kcal)	1124	246	955	331	1108	147	1042	316	1037	164	1010	387	NS	NS	NS
Lipid (kcal)	709	262	671	194	699	203	540	186	739	378	521	207	NS	NS	NS
Protein (kcal)	325	109	304	53	319	82	322	70	315	120	281	107	NS	NS	NS

Note: Lean women with percent body fat of 29% or lower (n=9).
HA (high adiposity) women with percent body fat of 29.1% or higher (n=8).

The possible differences in the measured and reported EI values were also investigated, since two different methods of measuring energy and macronutrient intakes were employed in this study. **Figure 6** compares the EI values measured inside the laboratory and reported by participants with 3-day food journals across the menstrual cycle according to body fat percentage. A significant interaction was found between the method used (direct measurement vs. food journal) and body fat percentage ($p<0.05$), where women with a higher adiposity level had a lower reported EI when compared to their EI values measured inside the laboratory. In addition, no correlations were noted between measured and reported EI values in all participants for each phase of the menstrual cycle.



Note: 1, Early Follicular; 2, Late follicular/ovulation; 3, Mid-Luteal.

Lean women with percent body fat of 29% or lower (n=9).

HA (high adiposity) women with percent body fat of 29.1% or higher (n=8).

Figure 6. Comparison of energy intake values measured inside the laboratory and reported with 3-day food journals according to body fat percentage.

4.4 RESTING METABOLIC RATE AND PHYSICAL ACTIVITY ENERGY EXPENDITURE

As shown in **Table 7**, no significant differences were noted in RMR, PAEE (kcal) and the amount of daily minutes spent doing sedentary, light, moderate and vigorous physical activities across the menstrual cycle in lean and HA women. No significant differences between groups, and interactions between menstrual cycle phase and body fat percentage were also noted when investigating all measured values of EE. Additionally, when controlling for the length of time for which each participant wore the accelerometer (i.e. data presented in mean daily kcal from PAEE/number hours wearing the accelerometer), no significant differences in total EE (kcal) were noted across phases (data not shown). Finally, certain correlations were noted during the late follicular/ovulation phase only; a positive correlation was found between percent body fat and the amount of daily minutes spent doing sedentary activities ($r=0.521$; $p<0.05$), as well as a negative correlation between percent body fat and the amount of daily minutes spent doing moderate ($r=-0.516$; $p<0.05$) and vigorous physical activity ($r=-0.578$; $p<0.05$).

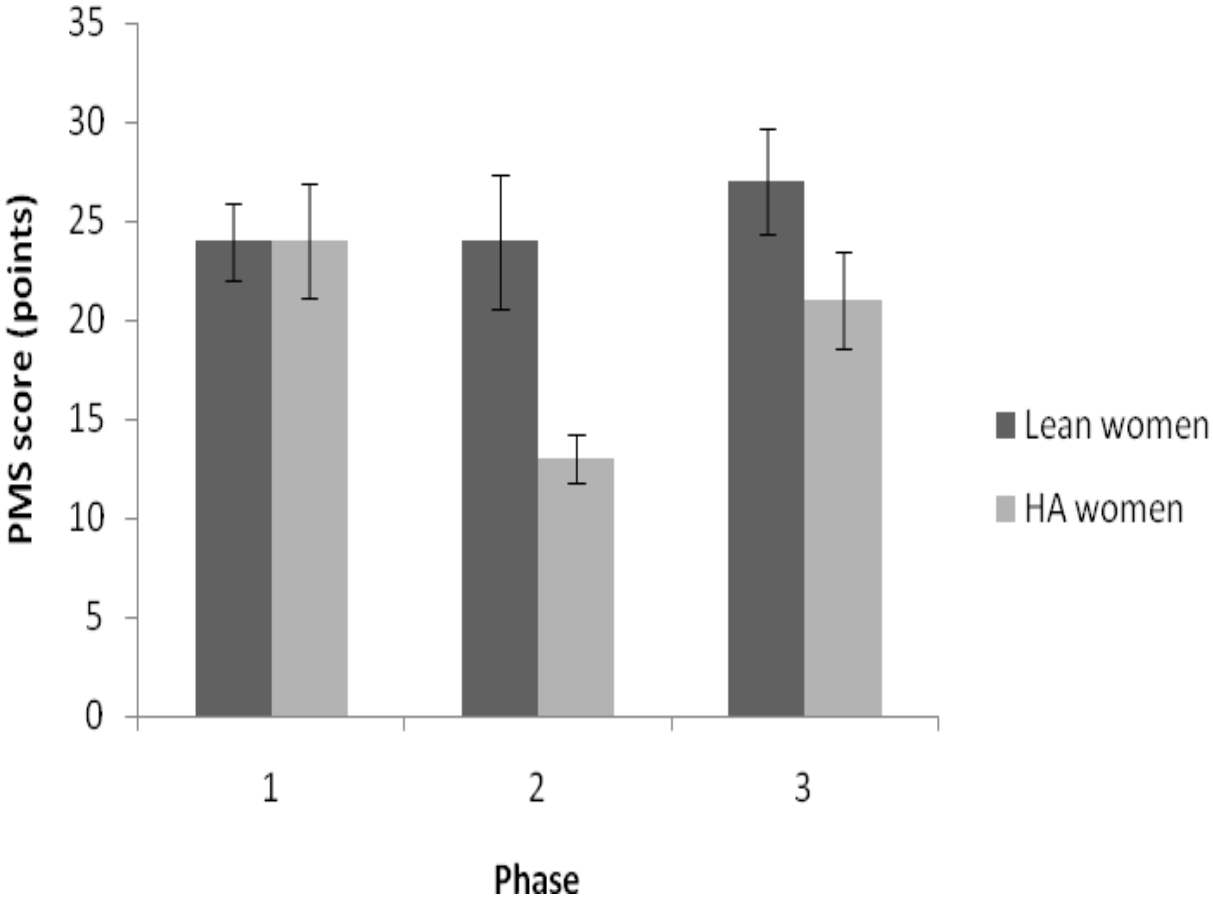
Table 7. RMR, daily PAEE (kcal) and daily minutes spent doing sedentary, light, moderate and vigorous physical activities across the menstrual cycle according to percent body fat.

	Early Follicular				Late follicular/ovulation				Mid-Luteal						
	Lean		HA		Lean		HA		Lean		HA				
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Phase	Fat %	Phase* Fat %
RMR	1319	100	1325	137	1340	145	1332	127	1312	141	1313	122	NS	NS	NS
PAEE (kcal)	815	301	804	335	840	244	852	269	857	295	857	340	NS	NS	NS
Sedentary (mins)	958	126	958	157	901	111	958	105	906	118	957	139	NS	NS	NS
Light (mins)	238	38	246	93	256	58	219	35	261	69	233	53	NS	NS	NS
Moderate (mins)	223	57	186	83	233	64	206	67	226	79	194	87	NS	NS	NS
Vigorous (mins)	19	18	13	9	19	19	14	10	21	16	18	15	NS	NS	NS

Note: Lean women with percent body fat of 29% or lower (n=9).
HA (high adiposity) women with percent body fat of 29.1% or higher (n=8).

4.5 THE OCCURRENCE AND SEVERITY OF PMS

A significant difference in the severity of PMS symptoms was noted across the menstrual cycle (25±10, 19±11, 25±10 points; $p<0.05$) in all participants. More specifically, there is a significant difference in the occurrence and severity of PMS symptoms between the late follicular/ovulation and mid-luteal phases ($p<0.01$), as well as a trend between the early follicular and late follicular/ovulation phases ($p=0.07$) in all participants. No significant difference between groups were noted; however, a significant interaction was found between the severity of PMS across the menstrual cycle and percent body fat ($p<0.05$). As shown in **Figure 7**, the occurrence and severity of PMS symptoms decrease during the late follicular/ovulation phase in women with higher adiposity levels but remains relatively stable in lean women, in comparison to the early follicular phase. Finally, no correlations were noted between all aspects of PAEE, energy and macronutrient intakes and percent body fat with the severity of PMS.



Note: 1, Early Follicular; 2, Late follicular/ovulation; 3, Mid-Luteal.

Lean women with percent body fat of 29% or lower (n=9).

HA (high adiposity) women with percent body fat of 29.1% or higher (n=8).

PMS scores for lean women: (24±8, 24±14, 27±11 points).

PMS scores for HA women: (26±12, 14±5, 22±10 points).

Figure 7. Points obtained on the shortened PMS assessment form across the menstrual cycle according to percent body fat.

4.6 THREE FACTOR EATING QUESTIONNAIRE

No significant differences were noted between dietary restraint (8±4, 8±5, 8±4 points; p=NS), dishinibition (5±3, 5±3, 5±3 points; p=NS) and perceived hunger (6±3, 6±4, 6±4 points; p=NS) scores across the menstrual cycle in all participants. Moreover, no significant differences between groups, as well as interactions between dietary restraint, dishinibition and perceived hunger scores in relation to percent body fat across the cycle were noted (data not shown). Finally, no correlations were noted between dietary restraint, dishinibition and perceived hunger scores, and energy and macronutrient intakes, the occurrence and severity of PMS, as well as percent body fat (data not shown).

4.7 SENSITVITY TO REWARD AND THE RELATIVE REINFORCING VALUE OF FOOD

No significant difference was noted in sensitivity to reward scores between groups (15±4 vs. 14±3 points; p=NS). As for food reinforcement, no significant differences were found in snack points, fruit points, snack button presses, fruit button presses and percentage of snack points earned across the menstrual cycle in lean and HA women (**Table 8**). In addition, no significant differences between groups, and no significant interactions were noted between percent body fat and all RRV of preferred food measurements across the menstrual cycle. However, positive correlations were noted between body fat percentage and fruit points during the late follicular/ovulation ($r=0.621$, $p<0.01$) and mid-luteal phases ($r=0.696$, $p<0.01$). Negative correlations were also noted between body fat percentage and snack points in all phases (data not shown). Similarly, positive correlations were noted between body fat percentage and fruit button presses during the early follicular ($r=0.550$,

$p<0.05$), late follicular/ovulation ($r=0.652$, $p<0.01$) and mid-luteal ($r=0.684$, $p<0.01$) phases. Negative correlations were also noted between body fat percentage and snack button presses in all phases (data not shown).

Table 8. Relative reinforcing value of food computer task results and preferred snack, fruit and combined snack/fruit intakes across the menstrual cycle according to percent body fat.

	Early Follicular				Late follicular/ovulation				Mid-Luteal						
	Lean		HA		Lean		HA		Lean		HA				
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Phase	Fat %	Phase* Fat %
Snack points	8	7	8	4	11	6	7	4	12	5	9	4	NS	NS	NS
Fruit points	12	7	12	4	9	6	13	4	8	5	11	4	NS	NS	NS
Snack button presses	96	82	81	51	138	81	82	57	145	67	106	49	NS	NS	NS
Fruit button presses	73	40	81	21	54	38	81	24	51	31	70	23	NS	NS	NS
% Snack points	40.6	33.3	38.1	19.1	55.0	30.4	36.3	20.5	59.4	23.9	44.4	19.5	NS	NS	NS
Snack intake (kcal)	106	103	100	149	95	51	104	113	163	120	170	174	0.06	NS	NS
Fruit intake (kcal)	57	49	61	56	47	39	57	13	59	37	57	44	NS	NS	NS
snack/fruit intake(kcal)	162	103	161	178	141	63	161	114	223	127	227	172	0.06	NS	NS

Note: Lean women with percent body fat of 29% or lower (n=9).

HA (high adiposity) women with percent body fat of 29.1% or higher (n=8).

Preferred snack and fruit intake were also measured. These represent the amount (kcal) of the participants' preferred snacks and fruits consumed that were earned based on their scores obtained on the RRV of preferred food task. It is also important to note that no significant difference in total EI prior to completing the RRV of preferred food task across the menstrual cycle was noted (data not shown), suggesting that *ad libitum* access to foods prior to doing this task seems to not have not influenced the participants' responses on the task, as well as preferred food intake following the task. An so, with regard to preferred food intakes, a trend was noted for preferred snack intake and combined preferred snack and fruit intakes across the menstrual cycle in all participants (**Table 8**). However, no significant difference was noted when looking at preferred fruit intake across the menstrual cycle. No significant differences and interactions were noted between snack, fruit and total preferred food intakes and body fat percentage across the cycle. However, positive correlations were noted between preferred fruit intake and fruit points during the early follicular ($r=0.510$, $p<0.05$), late follicular/ovulation ($r=0.647$, $p<0.01$) and mid-luteal ($r=0.730$, $p<0.01$) phases. On the contrary, negative correlations were noted between preferred fruit intake and snack points during each phase of the menstrual cycle (data not shown). Positive correlations were also noted between preferred fruit intake and fruit button presses, but only during the late follicular/ovulation ($r=0.620$, $p<0.01$) and the mid-luteal ($r=0.688$, $p<0.01$) phases. Similarly, during these two same phases, negative correlations were noted between fruit intake and snack button presses (data not shown). As for the early follicular phase, positive correlations were actually noted between preferred snack intake and snack points ($r=0.510$, $p<0.05$), as well as snack button presses ($r=0.534$, $p<0.05$). Similarly, during the early

follicular phase only, negative correlations were noted between snack intake and fruit points and fruit button presses (data not shown).

CHAPTER 5 - DISCUSSION

The primary aim of this study was to evaluate the variations in EI, macronutrient intake and EE, across the different phases of the menstrual cycle in women with different adiposity levels. The results of the present study showed no significant differences in total energy, carbohydrate, lipid and protein intakes across the menstrual cycle in lean women and in women with a higher adiposity level, rejecting the first hypothesis. In regard to RMR values, this hypothesis was supported since no significant changes were noted across the menstrual cycle in both groups. As for PAEE, no significant variations in total PAEE (kcal) or time spent doing sedentary, light, moderate and vigorous physical activities were noted across the menstrual cycle in both groups, rejecting this hypothesis.

This study also aimed to evaluate secondary factors, such as variations in female sex-steroid hormones, leptin, the occurrence and severity of PMS and food reinforcement. In regard to sex-steroid hormones, FSH, LH, estrogen and progesterone levels followed a normal pattern across the menstrual cycle; FSH and LH levels attained a peak during the late follicular/ovulation phase, estrogen levels increased with each phase of the cycle and progesterone levels were highest during the mid-luteal phase. Moreover, no significant difference between groups and no significant interaction were noted between body fat percentage and each of these hormones across the menstrual cycle, suggesting that adiposity level does not affect sex-steroid hormone levels, which supports this hypothesis. As for leptin, no significant variations in this hormone were noted across the menstrual cycle. This rejects the hypothesis which stated that leptin levels would be significantly higher during the mid-luteal phase when compared to both the early follicular and late follicular/ovulations

phases. However, as expected, the HA women did have higher levels of leptin. The results of the present study did show significant differences in PMS scores across the menstrual cycle. A significant interaction was also noted between PMS scores and body fat percentage, which surprisingly showed that lean women scored higher than the HA women on the shortened PMS assessment form during the late follicular/ovulation and the mid-luteal phases. This rejects the hypothesis which stated that women with higher adiposity levels will have higher scores on this questionnaire during the mid-luteal phase. Finally, no significant differences were noted in snack and fruit points, snack and fruit button presses and percentage of snack points earned in both groups across the menstrual cycle. Additionally, no significant differences between groups and no significant interactions were noted between these factors of food reinforcement and body fat percentage across the menstrual cycle. This rejects the hypothesis stating that women with high adiposity levels will show higher food reinforcement for snack foods.

5.1 ASSESSING PROPER TESTING TIMES: MEASURES OF BASAL TEMPERATURE AND SEX-STEROID HORMONES

First and foremost, no significant variations were noted in basal temperature across the menstrual cycle. However, basal temperatures did slightly increase across the menstrual cycle, suggesting that ovulation did occur and that testing was done during the proper phase of the menstrual cycle. In order to better evaluate the occurrence of ovulation and properly determine each phase, female sex-steroid hormone levels were evaluated. Significant differences in FSH, LH, estrogen and progesterone levels were noted across the menstrual cycle; higher levels of FSH and LH were noted during the late follicular/ovulation phase,

while estrogen levels increased with each phase and progesterone was at its highest level during the mid-luteal phase. The high levels of FSH and LH during the late follicular/ovulation phase is suggestive that ovulation did occur for each participant. Moreover, increased estrogen levels during the late follicular/ovulation and the mid-luteal phases, as well as higher levels of progesterone during the mid-luteal phase, suggest that these hormones followed a normal variation pattern across the menstrual cycle. All participants' FSH, LH, estrogen and progesterone levels were also within the normal, recommended range (Bermant & Davidson, 1974) for each phase. However, one participant was excluded from the analyses due to very low measured sex-steroid hormone levels across her menstrual cycle, which suggested that her cycle was anovulatory.

5.2 ANTHROPOMETRY

The results of the present study show that there were no significant variations in body weight and body fat percentage across the menstrual cycle in lean and HA women. As expected, the HA women showed significantly higher values for body weight, body fat percentage and fat mass, while a trend was noted for BMI, in comparison to the lean women. With no significant differences in fat-free mass between groups, it is possible to conclude that the higher body weight values noted in the HA women are due to higher levels of fat mass and naturally, body fat percentage. Additionally, a significant difference in fat-free mass was noted between the early follicular and late follicular/ovulation phases for all participants. This difference may simply be due to increases in water retention during the early follicular phase (Johnson et al., 1995) rather than actual changes in muscle mass, especially since no significant difference in fat mass was noted across the menstrual cycle in

all participants. The drop in progesterone levels prior to the start of menses has shown to increase water and salt retention (Michell, 1975; Frye & Demolar, 1993), thus leading to an increase in fluid retention at this time.

It is also important to note that the average BMI values for both groups are within the normal recommended range, while the average values for body fat percentage for each group are within the "normal range" for lean women and within the "overweight range" for the women with higher adiposity levels (Kennedy, Shea, & Sun, 2009), indicating that BMI may not always be an accurate indication of body adiposity or obesity. A novel aspect of the present study is that body composition, instead of BMI, was the primary consideration when looking at the variations in energy balance across the menstrual cycle.

5.3 ENERGY INTAKE AND ENERGY EXPENDITURE

By directly assessing energy and macronutrient intakes inside the laboratory during three distinct phases of the menstrual cycle, no significant differences were noted in energy and macronutrient intakes across the menstrual cycle in all participants. Along those lines, a mean difference in total EI of only 89 and 39 kcal were noted between the early follicular and the late follicular/ovulation phases, as well as between the late follicular/ovulation and the mid-luteal phases, respectively. No significant differences in energy and macronutrient intakes were also noted across the menstrual cycle through the use of 3-day food journals, indicating that the reported food journal results under free-living conditions and following each experimental session are comparable to what was directly measured inside the

laboratory when assessing energy and macronutrient intakes across menstrual cycle phase. Most studies which have previously evaluated the possible variations in energy and macronutrient intakes across the menstrual cycle have noted significantly higher EI (Li et al., 1999; Johnson et al., 1994; Dalvit et al., 1981; Lissner et al., 1988; Lyons et al., 1989; Dalvit-McPhillips, 1983; Gong et al., 1989; Manocha et al., 1986; Maritini et al., 1994; Tarasuk & Beaton, 1991), lipid (Li et al., 1999; Johnson et al., 1994; Martini et al., 1994; Tarasuk & Beaton, 1991) and carbohydrate (Li et al., 1999; Dalvit-McPhillips, 1983) intakes during the luteal phase. However, the use of different methodologies and testing times and/or frequencies in order to assess EI may explain the divergence in the results obtained by these studies. With increases in EI ranging from 87-500 kcal during the luteal phase in comparison to the follicular phase (**Table 1**), most studies (Li et al., 1999; Dalvit et al., 1981; Dalvit-McPhillips, 1983; Manocha et al., 1986; Maritni et al., 1994) which have noted higher variations in EI have employed dietary recall methods or food journals, and have measured this variable on two separate occasions (follicular and luteal phases). On the other hand, studies which have directly measured EI inside the laboratory found smaller variations in the latter across the menstrual cycle (Lissner et al., 1988; Fong & Kretsch, 1993). For instance, Lissner et al. (1988) noted a significant increase of 87 kcal in the luteal phase when compared to the follicular phase. Fong and Kretsch (1993) also directly measured *ad libitum* energy and macronutrient intakes inside the laboratory setting during four phases of the menstrual cycle (menses, follicular, ovulation and luteal) in 9 lean women, noting a trend in carbohydrate intake but no significant differences in total EI across the menstrual cycle.

In the present study, the direct assessment of energy and macronutrient intakes inside the laboratory through the use of a food menu (McNeil et al., 2011) may potentially present more accurate results than those reported with food journals under free living conditions, especially when assessing food intake in overweight or obese individuals. When comparing the measured and reported energy and macronutrient intakes, the lean individuals reported similar calorie intakes using a food journal in comparison to what they consumed inside the laboratory. However, the HA women reported having consumed approximately 400-500 calories less, when compared to their EI directly measured inside the laboratory. Past studies have shown that under-reporting and/or under-consumption in food journals are more common in both women (Black et al., 1991) and in overweight/obese individuals (Lissner, Heitmann, & Lindroos, 1998; Prentice et al., 1986) when assessed through the calculation of EI/estimated basal metabolic rate ratio and total EE by doubly labelled water, respectively. And so, it is possible that under-reporting and/or under-consumption may in part explain the lower reported values of EI in the HA women.

With regard to measurements of EE, no significant differences were noted in the RMR of all participants across the menstrual cycle. There seem to be conflicting results on this topic in the literature; increases in total EE and RMR seem to occur following ovulation when progesterone levels are high (Solomon et al., 1982; Webb, 1986) but these increases did not occur in all participants. Even though found to be non-significant, in the present study, the highest RMR values were noted during the late follicular/ovulation phase in both lean and HA women. And so, the slightly higher values in EI noted during this same phase may possibly lead to a slight increase in RMR, hence affecting total EE, or vice versa.

However, even though these two parameters showed similar variations across the menstrual cycle, no correlations were noted between RMR, PAEE and total energy and macronutrient intakes in each phase. Indeed, the non-significant changes in RMR may naturally be related to the slight variations in body weight and fat percentage (Bandini, Must, Phillips, Naumova, & Dietz, 2004) noted across the menstrual cycle. However, when comparing RMR values between groups, no significant differences were noted between lean and HA women, even though a significant difference in body weight was noted between these groups. This may be explained by the non-significant differences in fat-free mass noted between groups. Fat-free mass has been suggested to be one of the best predictors of RMR values (Ravussin, Burnand, Schutz, & Jécquier, 1982). Studies (James, Dauncey, & Davies, 1978; Halliday et al., 1979) have also shown that higher RMR values noted in overweight and obese individuals, in comparison to lean women, are most often related to an increase in absolute fat-free mass vs. increased fat mass. More specifically, an increase in 1 kg of fat mass leads to an increase of 4.5 kcal in RMR, while an increase in 1 kg of fat-free mass induces an increase of 13 kcal in RMR values (Elia, 1992).

As for daily PAEE, no significant differences were noted in the latter, as well as in the amount of daily time spent doing sedentary, light, moderate and vigorous physical activities across the menstrual cycle through the use of accelerometers. These results are similar to what has been previously noted through the use of physical activity journals in 26 lean women (Johnson et al., 1994). Similar values in PAEE (kcal) were also noted between groups. However, it is important to note that the lean participants spent more time doing

moderate and vigorous type activities, even though they may have expended a similar amount of kcal in comparison to the HA participants.

In summary, these slight, but not significant, variations in energy balance may suggest that the menstrual cycle may not be of practical concern when designing a food assessment study. Furthermore, it may be safe to conclude that these slight variations in energy balance are in accordance with the slight, but not significant, changes in body weight and body fat percentage seen across the menstrual cycle in both lean and HA women.

5.4 SECONDARY FACTORS: LEPTIN LEVELS, THE OCCURRENCE AND SEVERITY OF PMS AND THE RELATIVE REINFORCING VALUE OF PREFERRED FOODS

No significant variation in leptin levels was noted across the menstrual cycle in all participants. Even though many studies (Hardie et al., 1997; Riad-Gabriel et al., 1998; Al-Harithy et al., 2006; Mannucci et al., 1998; Thong et al., 2000) have noted a significant variation pattern in leptin levels across the menstrual cycle, other studies (Capobianco et al., 2010; Mills et al., 1998; Teirmaa et al., 1998) have noted no significant variation in leptin levels across the menstrual cycle, which is in agreement with our results. The different results obtained by different studies may be in part due to the frequency at which measurements of leptin levels were taken. The studies which have measured leptin levels on 4 or more occasions (Mannucci et al., 1998; Al-Harithy et al., 2006; Riad-Gabriel et al., 1998; Hardie et al., 1997) across the menstrual cycle were able to note significant differences. Many of these studies (Mannucci et al., 1998; Riad-Gabriel et al., 1998; Hardie et al., 1997), even though presenting leptin results based on menstrual cycle phase, measured

leptin from blood samples taken every 2-3 days for one entire menstrual cycle. On the other hand, the present study, as well as other studies which have noted no significant variation in leptin concentrations across the menstrual cycle (Capobianco et al., 2010; Mills et al., 1998; Teirmaa et al., 1998), have only measured leptin on three occasions across the menstrual cycle, suggesting that frequent measurements of leptin may be needed to note significant variations in this hormone across the menstrual cycle.

The occurrence and severity of PMS symptoms were evaluated through the shortened premenstrual assessment form during each phase. Based on these results, PMS symptoms seem to be less severe during the late follicular/ovulation phase in all participants in comparison to the early follicular and mid-luteal phases, which is in accordance with other studies (Johnson et al., 1995; Cross et al., 2001; Both-Orthman et al., 1988), which have noted higher prevalence of PMS during the late luteal phase and at the start of menses. However, a difference between groups is apparent; lean women reported more severe PMS symptoms during the mid-luteal phase, while PMS symptoms seemed to be more predominant during the early follicular phase in HA women. In addition, the HA women reported less severe PMS symptoms during the late follicular/ovulation phase and the mid-luteal phase in comparison to their lean counterparts. These results are not in accordance with what Masho et al. (2005) had previously reported in a cohort of 874 women aged between 18-44 years, suggesting that the prevalence of PMS increased with BMI, meaning that overweight and obese women were more likely to suffer from PMS symptoms in comparison to lean women. Even though the number of participants in the present study who are classified as being obese according to body fat percentage (n=2) is very limited, the exact

reason behind the higher prevalence in PMS in the lean participants during the late follicular/ovulation and the mid-luteal phases is unclear, especially since lean women, despite reporting more severe symptoms of PMS, did not necessarily have a higher total EI. However, in a study done by Bryant et al. (2006), no significant differences in EI were noted during the follicular and luteal phases between women who reported suffering from PMS as compared to a group of controls who reported not suffering from PMS. In addition, even though not significant, the women who reported suffering from PMS surprisingly consumed more calories during the follicular phase, which is similar to the findings of the current study even though PMS symptoms were not necessarily reported being higher at this time. Moreover, the questionnaire itself, being a shortened version of the original 95-item premenstrual assessment form, has been shown to put more emphasis on the physical symptoms of PMS; the reduced number of items on the questionnaire may not be able to record the full range of symptoms which constitute PMS (Haywood, Slade, & King, 2002). Following this further, it may be possible that many physical symptoms related to PMS (e.i. pain and tenderness, bloating, weight gain) may actually lead to a decrease in EI, while the increase in EI may be seen most often by PMS sufferers who experience more emotional symptoms such as mood swings, irritability and stress. It has been previously shown that PMS symptoms related to negative mood, such as tension, anger, depression and tiredness are most often related to increases in food cravings for high dense, palatable foods (Rogers & Smit, 2000; Wurtman & Wurtman, 1989). Based on the current findings, it seems that the occurrence and severity of PMS symptoms may actually not be responsible for higher total EI. However, it could be hypothesized that the slightly lower EI results noted during the

early follicular and mid-luteal phases of the menstrual cycle in both groups may possibly be related to physical, rather than emotional, PMS symptoms reported by these participants.

The present study is one of the first to measure the RRV of preferred foods across the menstrual cycle and relate these results to the different components of energy balance. Even though no significant differences were noted in snack and fruit points, snack and fruit button presses and percentage of snack points, it is very interesting to consider that trends were noted in preferred snack and combined preferred snack/fruit intakes across the menstrual cycle, with all participants consuming on average slightly more calories from preferred foods during the mid-luteal phase. It is also interesting to note that the point and button press distributions according to preferred food type for all phases were generally accurate predictors of preferred food intakes, in which case higher snack points and snack button presses were generally associated with higher preferred snack intake for instance. And so, the participants in the present study consumed a slightly larger quantity of their preferred foods during the mid-luteal phase, represented by a trend noted across menstrual cycle phase, even though total EI was not necessarily higher during this phase, as opposed to what many other studies have previously reported (**Table 1**). This near significant increase in EI from combined preferred fruit/snack foods during the mid-luteal phase may most likely be attributed to an increase in snack food intake during this phase, since preferred snack foods contain on average more calories per serving vs. preferred fruits. Moreover, the near significant increase in snack intake during the mid-luteal phase in comparison to the non-significant differences in fruit intake across the menstrual cycle may also suggest that increases in preferred high dense, palatable foods is most often present during the mid-luteal

phase of the menstrual cycle. However, even though a slight increase in the consumption of preferred high dense palatable foods seems to occur during the mid-luteal phase, no significant increase in total EI was noted during this phase of the menstrual cycle, suggesting that the participants may have compensated by decreasing their intake of other foods, such as meal-type foods for instance.

When looking at the results obtained by the lean and HA women, it is interesting to note that a positive correlation was noted between body fat percentage and fruit points and fruit button presses for all phases, while a negative correlation was noted between body fat percentage and snack points and snack button presses, suggesting that the lean women obtained more snack points and snack button presses for all phases, while the opposite occurred in the HA women. Moreover, the lean women also consumed a larger quantity of snack foods during each phase, in comparison to the HA women, even though these differences were found to be non-significant. Based on these findings, it may be suggested that the HA women in the present study may have found the snack foods less reinforcing and consumed a slightly smaller quantity of this preferred food because of the effort needed to obtain the latter, in comparison to their lean counterparts. Certain studies have noted a decrease in preferred food reinforcement (Smith & Epstein, 1991) and food intake (Reznich & Balch, 1977) in overweight and obese individuals when the number of button presses or the amount of effort required to obtain a preferred food increased over time. It was also noted that lean individuals' eating choices were not affected by the amount of effort required to obtain a food reward (Reznich & Balch, 1977). And so, the slightly lower values in preferred snack reinforcement and preferred snack intake in the HA women, while

maintaining a slightly higher overall total EI, may be related to the amount of work required to obtain the preferred foods; a factor which seems to not affect the choices in preferred snack reinforcement and preferred snack food intake in lean women.

In the same way, even though lean women consumed a slightly higher quantity of preferred snack foods, they did not necessarily have an overall higher EI in comparison to the HA women. It may thus be possible that lowered post-meal satisfaction (Tucci, Murphy, Boyland, Dye, & Halford, 2010) related to food intake may be an important driving factor in explaining why total EI was slightly higher in HA women, even though they consumed a similar amount of calories from preferred foods as did the lean women. It has been shown that the consumption of a favourite meal is associated with dopamine release (Pannacciulli et al., 2006). It has also been shown that obese individuals have lower density of dopamine receptors (Wang et al., 2001), suggesting that the reward centers in the brain in these individuals may not be as sensitive to food intake; thus leading to possible overconsumption as a means to compensate for the decreased activation of the reward centers of the brain modulated by dopamine (Berridge & Robinson, 1998). And so, the consumption of preferred foods would lead to normal dopamine releases in lean women, providing an adequate level of satisfaction and possibly leading to a decrease in EI following the consumption of these preferred foods. However, in the case of women with higher adiposity levels, it may be hypothesized that the consumption of these preferred foods may not provide the same level of satisfaction related to reward, thus leading to higher consumptions of other foods throughout the day.

In summary, even though non-significant differences were noted in snack food reinforcement, preferred snack intake and total EI between groups, it is possible that both environmental constraints and physiological factors may explain the slightly lower preferred snack food reinforcement and preferred snack intakes in the HA women while still maintaining a slightly higher total EI, in comparison to the lean women. It is also interesting to note that the lean women in this study seem to suffer from more severe PMS symptoms, as well as work slightly harder for snack food points. It could thus be hypothesized that the occurrence and severity of PMS, especially during the mid-luteal phase, in lean women may be related to an increase in preferred snack reinforcement and preferred snack food intake. However, no correlation was noted between the severity of PMS and the RRV of preferred snack food scores and snack food intake for all phases of the menstrual cycle in all participants.

All things considered, the present study rejected two of the primary hypotheses, since no significant differences in EI, macronutrient intake and PAEE were noted across the menstrual cycle, as well as between groups. No significant difference in RMR values were also noted across the menstrual cycle and between groups, in this case accepting our initial hypothesis. As for secondary hypotheses, no significant differences were noted in sex-steroid hormone levels, accepting this hypothesis. However, no significant differences in leptin levels were noted across the menstrual cycle, while a significant difference in leptin was noted between groups; in part rejecting this hypothesis. Finally, both hypotheses related to the occurrence and severity of PMS and food reinforcement were not supported by results since these factors were not predominant in HA women. Even though most of the hypotheses

were rejected, the results of this study does provide a new insight into the slight, but non-significant, variations in the energy balance which may occur across the menstrual cycle. Furthermore, the negative correlation between snack reinforcement and leptin levels, the higher prevalence and severity of PMS, as well as the trends for higher preferred snack and total preferred food intakes, noted during the mid-luteal phase may indeed affect different aspects of EI, while not necessarily increasing total food consumption during this phase.

CHAPTER 6 - LIMITATIONS AND FUTURE PERSPECTIVES

Even though this study does present many methodological strengths by directly measuring energy and macronutrient intakes inside the laboratory and more accurately measuring PAEE through the use of accelerometers, as well as determining the phase of the menstrual cycle with measures of body temperature and female sex-steroid hormone levels; certain limitations were present and inevitable. First and foremost, if the sample size would have been larger, it is possible that certain trends, such as preferred snack and combined snack/fruit intakes, across the different menstrual cycle phases may have approached significance. As for the main outcomes of this study, the values for power were very low given that measured EI and macronutrient intakes were almost the same across the menstrual cycle (**Table 5**). Indeed, the power was 0.09, 0.07, 0.10 and 0.13 for energy, carbohydrate, lipid and protein intakes respectively. This was also the case for RMR and PAEE values (**Table 7**), where the power was 0.12 and 0.11 for RMR and PAEE respectively. However, there was no clear evidence for a trend indicating a difference for total EI, macronutrient intake, RMR and PAEE across the menstrual cycle in all participants. In fact, the estimate of effect size was extremely low for these analyses; the estimate effect size for energy, carbohydrate, lipid and protein intakes, RMR and PAEE being 0.02, 0.01, 0.02, 0.03, 0.03 and 0.03 respectively. Similar power and estimate of effect size values were noted for between-subject analyses in regard to the main outcomes of this study. The values for power and estimate of effect size between groups were 0.23 and 0.10 for total EI, 0.22 and 0.09 for carbohydrate intake, 0.16 and 0.06 for lipid intake, 0.19 and 0.08 for protein intake, 0.05 and 0.00 for RMR, 0.05 and 0.00 for PAEE. This means that increasing the number of subjects would have very likely lead to the same results for our main outcomes. So, increasing the

number of subjects any further would have very likely resulted in the same outcome, at least as far as EI, macronutrient intake, RMR and PAEE are concerned.

Participants were also not intentionally randomized. Four participants had their first session during the early follicular phase, while eight and seven had their first sessions during the late follicular/ovulation mid-luteal phases respectively. And so, even though not all participants had their first testing session during the same phase, this randomization was not done intentionally.

In addition, normal day to day variations in EI could have occurred and affected the results since direct measurements of EI were only taken for 1 day (8h00-17h30) inside the laboratory for each phase. Evening snacking was also not measured on these days due to each experimental testing session ending at 17h30. It is thus recommended for future studies to have the participants note what they consumed in the evening outside the laboratory, following each in-laboratory session, which would provide information on the amount of food and beverages they would consume that same day once they have left the laboratory. Similarly, limiting each participant's normal activity patterns by asking them to remain in a room for close to 10 hours of the day might have also influenced their eating patterns. Presenting a copy of the food menu to the participants prior to the first day of testing would have given the participants a chance to get accustomed to the choices that will be offered to them during their testing sessions and may have helped in preventing possible occurrences of under- or overeating, and this especially during the 1st testing session.

Certain inevitable factors also presented limitations in the present study. For instance, not all participants had a 28-day cycle. Furthermore, there were unforeseeable variations noted in some of the participant's cycle lengths from one cycle to the next. For instance, the number of days counted in one's previous cycle did not necessarily correspond to the exact number of days of their following cycle. These slight variations in cycle length made it more difficult to accurately determine the time of ovulation and the exact length of every phase when establishing testing times. Different environmental factors not related to the menstrual cycle (i.e. stress, exams, recently moving to a new city) could have also influenced cycle length and different aspects of EI and EE, such as eating habits and regular exercise routines. In future testing, establishing the time of ovulation and proper testing times may be facilitated by having the participants measure and take note of their basal temperature every morning for the length of one menstrual cycle, in addition to noting the length of the latter, prior to the start of testing. Additionally, further pre-experimental screening may be required in order to exclude or postpone testing times of participants who may be experiencing higher than normal levels of stress due to different factors, such as recently moving to a new city and writing exams.

There was also a limitation with regard to the RRV of food task, in which case giving the participants *ad libitum* access to foods prior to the task may have influenced their scores on the RRV of food task if one of the preferred foods was offered on the menu and the participant consumed some of this food prior to doing the test. More specifically, six, seven and six participants consumed a preferred food or high dense snack prior to doing this task during the early follicular, late follicular/ovulation and mid-luteal phases, respectively.

Based on multivariate analyses, a significant difference in snack points earned and preferred snack foods consumed between groups (i.e. participants who consumed a preferred food or high dense snack prior to doing the RRV of food task vs. those who did not) was only noted during the early follicular phase. In future testing, it is suggested to provide the preferred foods towards the end of the day, or when the participant leaves the laboratory, in order to avoid the possible influence of preferred food consumption prior to completing the RRV of food task on the participant's answers on this test and preferred food intake hereafter.

Finally, only two obese individuals (classified according to body adiposity as having a body fat percentage of 39% or higher) took part in this study, meaning that the results obtained in this study should only be generalized to lean and overweight women, and not necessarily obese women. Moreover, when based on BMI alone, none of the participants had a BMI of 27 kg/m² or more, meaning than none of them were classified as being "overweight". The recruitment of overweight and obese women (classified according to body fat percentage) did increase in the latter part of recruitment, at which time both obese individuals and three overweight individuals were recruited. However, despite our best efforts, it was quite difficult to recruit an adequate number of obese individuals for this study.

The above limitations and difficulties encountered during this study would lead one to develop ideas and suggestions for future studies on this particular subject. First and foremost, it would be important to directly measure energy and macronutrient intakes over at least 2-3 days per menstrual cycle phase or one complete menstrual cycle in order to have a better idea of one's energy and macronutrient intakes over a longer period of time and to

minimize the effects of day to day variations in EI. The use of a “lunch box”, a new tool which has also been validated outside the laboratory with the food menu (McNeil et al., 2011), may be able to provide the necessary information in regard to energy and macronutrient intakes over 2-3 days per menstrual cycle phase.

It is also suggested to evaluate the occurrence of food cravings and the amount of kcal consumed which may be related to cravings or snacking across the menstrual cycle. Even though no significant differences were noted across the menstrual cycle in total energy and macronutrient intakes, there may be significant differences in the types of foods consumed by the participants. For instance, it would be interesting to evaluate if one may tend to consume more snack-type foods during one phase of the menstrual cycle vs. a different phase, thus obtaining most of their kcal from these types of foods vs. meal-type foods for example.

CHAPTER 7 - CONCLUSION

In conclusion, no significant differences were noted in EI, macronutrient intakes and EE across the menstrual cycle. The measurement of all these factors in the same individuals provides a better idea of the slight, but non-significant, variations in the energy balance which may occur across the menstrual cycle. In addition, the EI results in the present study are very interesting because they are not necessarily in accordance with past literature, which have mainly noted an increase in EI during the luteal phase when assessing EI through food journals and dietary recall methods. Similarly, the results obtained through the shortened PMS assessment form and the RRV of preferred food computer task presented unexpected findings, which provides a new insight into how these behavioral factors may affect different aspects of EI across the menstrual cycle in women with different adiposity levels. The slight, but not significant, variations in energy balance suggest that the menstrual cycle may not necessarily be of practical concern when designing a food assessment study. It is however warranted to directly measure energy and macronutrient intakes over the course of 2-3 days per menstrual cycle phase or one entire menstrual cycle, as well as look into the possible variations in the types of foods consumed (meal, snack, caloric beverage and water) in order to have a better idea of where these slight variations in EI may occur.

REFERENCES

Al-Harithy, R. N., Al-Doghaither, H., & Abualnaja, K. (2006). Correlation of leptin and sex hormones with endocrine changes in healthy Saudi women of different body weights. *Ann Saudi Med, 26,* 110-115.

Allen, S. S., McBride, C. M., & Pirie, P. L. (1991). The shortened premenstrual assessment form. *J Reprod Med, 36,* 769-772.

Angst, J., Sellaro, R., Merikangas, K. R., & Endicott, J. (2001). The epidemiology of perimenstrual psychological symptoms. *Acta Psychiatr Scand, 104,* 110-116.

Augustine, R. A., Ladyman, S. R., & Grattan, D. R. (2008). From feeding one to feeding many: hormone-induced changes in bodyweight homeostasis during pregnancy. *J Physiol, 586,* 387-397.

Bandini, L. G., Must, A., Phillips, S. M., Naumova, E. N., & Dietz, W. H. (2004). Relation of body mass index and body fatness to energy expenditure: longitudinal changes from preadolescence through adolescence. *Am J Clin Nutr, 80,* 1262-1269.

Bermant, G., & Davidson, J. M. (1974). The biological bases of sexual behavior New York:Harper Row

Berridge, K. C., & Robinson, T. E. (1998). What is the role of dopamine in reward: hedonic impact, reward learning, or incentive salience? *Brain Res Brain Res Rev, 28,* 309-369.

Bingham, S. A., Cassidy, A., Cole, T. J., Welch, A., Runswick, S. A., Black, A. E., Thurnham, D., Bates, C., Khaw, K. T., Key, T. J., et al. (1995). Validation of weighed records and other methods of dietary assessment using the 24 h urine nitrogen technique and other biological markers. *Br J Nutr, 73,* 531-550.

Black, A. E., Goldberg, G. R., Jebb, S. A., Livingstone, M. B., Cole, T. J., & Prentice, A. M. (1991). Critical evaluation of energy intake data using fundamental principles of energy physiology: 2. Evaluating the results of published surveys. *Eur J Clin Nutr, 45,* 583-599.

Both-Orthman, B., Rubinow, D. R., Hoban, M. C., Malley, J., & Grover, G. N. (1988). Menstrual cycle phase-related changes in appetite in patients with premenstrual syndrome and in control subjects. *Am J Psychiatry, 145,* 628-631.

Bryant, M., Truesdale, K. P., & Dye, L. (2006). Modest changes in dietary intake across the menstrual cycle: implications for food intake research. *Br J Nutr, 96,* 888-894.

Buffenstein, R., Poppitt, S. D., McDevitt, R. M., & Prentice, A. M. (1995). Food intake and the menstrual cycle: a retrospective analysis, with implications for appetite research. *Physiol Behav, 58,* 1067-1077.

Cameron, J., & Doucet, E. (2007). Getting to the bottom of feeding behaviour: who's on top? *Appl Physiol Nutr Metab, 32,* 177-189.

Capobianco, G., de Muro, P., Cherchi, G. M., Formato, M., Lepedda, A. J., Cigliano, A., Zinellu, E., Dessole, F., Gordini, L., & Dessole, S (2010). Plasma levels of C-reactive protein, leptin and glycosaminoglycans during spontaneous menstrual cycle: differences between ovulatory and anovulatory cycles. *Arch Gynecol Obstet, 282,* 207-213.

Chehab, F. F., Lim, M. E., & Lu, R. (1996). Correction of the sterility defect in homozygous obese female mice by treatment with the human recombinant leptin. *Nat Genet, 12,* 318-320.

Chehab, F. F., Mounzih, K., Lu, R., & Lim, M. E. (1997). Early onset of reproductive function in normal female mice treated with leptin. *Science, 275,* 88-90.

Cioffi, J. A., Van Blerkom, J., Antczak, M., Shafer, A., Wittmer, S., & Snodgrass, H. R. (1997). The expression of leptin and its receptors in pre-ovulatory human follicles. *Mol Hum Reprod, 3,* 467-472.

Cross, G. B., Marley, J., Miles, H., & Willson, K. (2001). Changes in nutrient intake during the menstrual cycle of overweight women with premenstrual syndrome. *Br J Nutr, 85,* 475-482.

Cunningham, M. J., Clifton, D. K., & Steiner, R. A. (1999). Leptin's actions on the reproductive axis: perspectives and mechanisms. *Biol Reprod, 60,* 216-222.

Czaja, J. A. (1978). Ovarian influence on primate food intake: Assessment of progesterone actions. *Physiol Behav, 21,* 923-928.

Czaja, J. A., & Goy, R. W. (1975). Ovarian hormones and food intake in female guinea pigs and rhesus monkeys. *Horm Behav, 6,* 329-349.

Dalvit, S. P. (1981). The effect of the menstrual cycle on patterns of food intake. *Am J Clin Nutr, 34,* 1811-1815.

Dalvit-McPhillips, S. P. (1983). The effect of the human menstrual cycle on nutrient intake. *Physiol Behav, 31,* 209-212.

Davidsen, L., Vistisen, B., & Astrup, A. (2007). Impact of the menstrual cycle on determinants of energy balance: a putative role in weight loss attempts. *Int J Obes (Lond), 31,* 1777-1785.

Dye, L., & Blundell, J. E. (1997). Menstrual cycle and appetite control: implications for weight regulation. *Hum Reprod, 12,* 1142-1151.

Dye, L., Warner, P., & Bancroft, J. (1995). Food craving during the menstrual cycle and its relationship to stress, happiness of relationship and depression; a preliminary enquiry. *J Affect Disord, 34,* 157-164.

Elia M. Organ and Tissue contribution to metabolic rate. In: Kinney JM, Tucker HN, eds. *Energy Metabolism: Tissue determinants and cellular corollaries. New York: Raven Press;* 1992: pp. 61-80.

Epstein, L. H., Bulik, C. M., Perkins, K. A., Caggiula, A. R., & Rodefer, J. (1991). Behavioral economic analysis of smoking: money and food as alternatives. *Pharmacol Biochem Behav, 38,* 715-721.

Evans, S. M., & Foltin, R. W. (2004). Pharmacokinetics of intravenous cocaine across the menstrual cycle in rhesus monkeys. *Neuropsychopharmacology, 29,* 1889-1900.

Farage, M. A., Neill, S., & MacLean, A. B. (2009). Physiological changes associated with the menstrual cycle: a review. *Obstet Gynecol Surv, 64,* 58-72.

Farage, M. A., Osborn, T. W., & MacLean, A. B. (2008). Cognitive, sensory, and emotional changes associated with the menstrual cycle: a review. *Arch Gynecol Obstet, 278,* 299-307.

Figlewicz, D. P., MacDonald Naleid, A., & Sipols, A. J. (2007). Modulation of food reward by adiposity signals. *Physiol Behav, 91,* 473-478.

Figlewicz, D. P., & Benoit, S. C. (2009). Insulin, leptin, and food reward: update 2008. *Am J Physiol Regul Integr Comp Physiol, 296,* R9-R19.

Fong, A. K., & Kretsch, M. J. (1993). Changes in dietary intake, urinary nitrogen, and urinary volume across the menstrual cycle. *Am J Clin Nutr, 57,* 43-46.

Frank, T. C., Kim, G. L., Krzemien, A., & Van Vugt, D. A. Effect of menstrual cycle phase on corticolimbic brain activation by visual food cues. *Brain Res, 1363,* 81-92.

Freeman, E. W., Schweizer, E., & Rickels, K. (1995). Personality factors in women with premenstrual syndrome. *Psychosom Med, 57,* 453-459.

Frye, C. A., & Demolar, G. L. (1994). Menstrual cycle and sex differences influence salt preference. *Physiol Behav, 55,* 193-197.

Gong, E. J., Garrel, D., & Calloway, D. H. (1989). Menstrual cycle and voluntary food intake. *Am J Clin Nutr, 49,* 252-258.

Goumenou, A. G., Matalliotakis, I. M., Koumantakis, G. E., & Panidis, D. K. (2003). The role of leptin in fertility. *Eur J Obstet Gynecol Reprod Biol, 106,* 118-124.

Groschl, M., Rauh, M., Dorr, H. G., Blum, W. F., Rascher, W., & Dotsch, J. (2002). Salivary leptin levels during the menstrual cycle and their relation to progesterone. *Fertil Steril, 77,* 1306-1307.

Halliday, D., Hesp, R., Stalley, S. F., Warwick, P., Altman, D. G., & Garrow, J. S. (1979). Resting metabolic rate, weight, surface area and body composition in obese women. *Int J Obes, 3,* 1-6.

Hardie, L., Trayhurn, P., Abramovich, D., & Fowler, P. (1997). Circulating leptin in women: a longitudinal study in the menstrual cycle and during pregnancy. *Clin Endocrinol (Oxf), 47,* 101-106.

Hartz, A., Barboriak, P., Wong, A., Katayama, K. P., & Rimm, A. A. (1979). The association of obesity with infertility and related menstrual abnormalities in women. *Int J Obes, 3,* 57-73.

Haywood, A., Slade, P., & King, H. (2002). Assessing the assessment measures for menstrual cycle symptoms: a guide for researchers and clinicians. *J Psychosom Res, 52,* 223-237.

Hess, D. L., & Resko, J. A. (1973). The effects of progesterone on the patterns of testosterone and estradiol concentrations in the systemic plasma of the female rhesus monkey during the intermenstrual period. *Endocrinology, 92,* 446-453.

Israel, S. L., & Schneller, O. (1950). The thermogenic property of progesterone. *Fertil Steril, 5,* 532-533.

James, W. P., Dauncey, M. J., & Davies, H. L. (1978). Elevated metabolic rates in obesity. *Lancet, 2,* 472.

Jas, P. E. 1996. Changes in food intake and mood across the menstrual cycle. Ph.D. thesis: University of Reading, UK.

Johnson, W. G., Carr-Nangle, R. E., & Bergeron, K. C. (1995). Macronutrient intake, eating habits, and exercise as moderators of menstrual distress in healthy women. *Psychosom Med, 57,* 324-330.

Johnson, W. G., Corrigan, S. A., Lemmon, C. R., Bergeron, K. B., & Crusco, A. H. (1994). Energy regulation over the menstrual cycle. *Physiol Behav, 56,* 523-527.

Julliard, A. K., Chaput, M. A., Apelbaum, A., Aime, P., Mahfouz, M., & Duchamp-Viret, P. (2007). Changes in rat olfactory detection performance induced by orexin and leptin mimicking fasting and satiation. *Behav Brain Res, 183,* 123-129.

Kennedy, A. P., Shea, J. L., & Sun, G. (2009). Comparison of the classification of obesity by BMI vs. dual-energy X-ray absorptiometry in the Newfoundland population. *Obesity (Silver Spring), 17,* 2094-2099.

Kitawaki, J., Koshiba, H., Ishihara, H., Kusuki, I., Tsukamoto, K., & Honjo, H. (2000). Expression of leptin receptor in human endometrium and fluctuation during the menstrual cycle. *J Clin Endocrinol Metab, 85,* 1946-1950.

Kopp, W., Blum, W. F., von Prittwitz, S., Ziegler, A., Lubbert, H., Emons, G., Herzog, W., Herpertz, S., Deter, H. C., Remschmidt, H., et al. (1997). Low leptin levels predict amenorrhea in underweight and eating disordered females. *Mol Psychiatry, 2,* 335-340.

Ladyman, S. R. (2008). Leptin resistance during pregnancy in the rat. *J Neuroendocrinol, 20,* 269-277.

Lappalainen, R., & Epstein, L. H. (1990). A behavioral economics analysis of food choice in humans. *Appetite, 14,* 81-93.

Li, E. T., Tsang, L. B., & Lui, S. S. (1999). Menstrual cycle and voluntary food intake in young Chinese women. *Appetite, 33,* 109-118.

Licinio, J., Negrao, A. B., Mantzoros, C., Kaklamani, V., Wong, M. L., Bongiorno, P. B., Mulla, A., Cearnal, L., Veldhuis, J. D., Flier, J. S., et al. (1998). Synchronicity of frequently sampled, 24-h concentrations of circulating leptin, luteinizing hormone, and estradiol in healthy women. *Proc Natl Acad Sci U S A, 95,* 2541-2546.

Lissner, L., Heitmann, B. L., & Lindroos, A. K. (1998). Measuring intake in free-living human subjects: a question of bias. *Proc Nutr Soc, 57,* 333-339.

Lissner, L., Stevens, J., Levitsky, D. A., Rasmussen, K. M., & Strupp, B. J. (1988). Variation in energy intake during the menstrual cycle: implications for food-intake research. *Am J Clin Nutr, 48,* 956-962.

Lyons, P. M., Truswell, A. S., Mira, M., Vizzard, J., & Abraham, S. F. (1989). Reduction of food intake in the ovulatory phase of the menstrual cycle. *Am J Clin Nutr, 49,* 1164-1168.

Mannucci, E., Ognibene, A., Becorpi, A., Cremasco, F., Pellegrini, S., Ottanelli, S., Rizzello, S. M., Massi, G., Messeri, G., & Rotella, C. M. (1998). Relationship between leptin and oestrogens in healthy women. *Eur J Endocrinol, 139,* 198-201.

Manocha, S., Choudhuri, G., & Tandon, B. N. (1986). A study of dietary intake in pre- and post-menstrual period. *Hum Nutr Appl Nutr, 40,* 213-216.

Martini, M. C., Lampe, J. W., Slavin, J. L., & Kurzer, M. S. (1994). Effect of the menstrual cycle on energy and nutrient intake. *Am J Clin Nutr, 60,* 895-899.

Masho, S. W., Adera, T., & South-Paul, J. (2005). Obesity as a risk factor for premenstrual syndrome. *J Psychosom Obstet Gynaecol, 26,* 33-39.

McNeil, J., Riou, ME., Razmjou, S., Cadieux, S., & Doucet, E. Validation of a food menu to measure energy and macronutrient intakes in a laboratory and under real-life conditions. *Br J of Nutr* (In revision).

Mela, D. J., & Rogers, P. J. (1998). Food, eating and obesity: The psychological basis of appetite and weight control London:Chapman and Hall

Michell, A. R. (1975). Changes of sodium appetite during the estrous cycle of sheep. *Physiol Behav, 14,* 223-226.

Michener, W., Rozin, P., Freeman, E., & Gale, L. (1999). The role of low progesterone and tension as triggers of perimenstrual chocolate and sweets craving: some negative experimental evidence. *Physiol Behav, 67,* 417-420.

Mills, P. J., Ziegler, M. G., & Morrison, T. A. (1998). Leptin is related to epinephrine levels but not reproductive hormone levels in cycling African-American and Caucasian women. *Life Sci, 63,* 617-623.

Murphy, K. G., & Bloom, S. R. (2004). Gut hormones in the control of appetite. *Exp Physiol, 89,* 507-516.

Pannacciulli, N., Del Parigi, A., Chen, K., Le, D. S., Reiman, E. M., & Tataranni, P. A. (2006). Brain abnormalities in human obesity: a voxel-based morphometric study. *Neuroimage, 31,* 1419-1425.

Pliner, P., & Fleming, A. S. (1983). Food intake, body weight, and sweetness preferences over the menstrual cycle in humans. *Physiol Behav, 30,* 663-666.

Prentice, A. M., Black, A. E., Coward, W. A., Davies, H. L., Goldberg, G. R., Murgatroyd, P. R., Ashford, J., Sawyer, M., & Whitehead, R. G. (1986). High levels of energy expenditure in obese women. *Br Med J (Clin Res Ed), 292,* 983-987.

Ravussin, E., Burnand, B., Schutz, Y., & Jequier, E. (1982). Twenty-four-hour energy expenditure and resting metabolic rate in obese, moderately obese, and control subjects. *Am J Clin Nutr, 35,* 566-573.

Reznick, H., & Balch, P. (1977). The effects of anxiety and response cost manipulations on the eating behavior of obese and normal-weight subjects. *Addict Behav, 2,* 219-225.

Riad-Gabriel, M. G., Jinagouda, S. D., Sharma, A., Boyadjian, R., & Saad, M. F. (1998). Changes in plasma leptin during the menstrual cycle. *Eur J Endocrinol, 139,* 528-531.

Rogers, P. J., & Smit, H. J. (2000). Food craving and food "addiction": a critical review of the evidence from a biopsychosocial perspective. *Pharmacol Biochem Behav, 66,* 3-14.

Rosenbaum, M., Nicolson, M., Hirsch, J., Heymsfield, S. B., Gallagher, D., Chu, F., & Leibel, R. L. (1996). Effects of gender, body composition, and menopause on plasma concentrations of leptin. *J Clin Endocrinol Metab, 81,* 3424-3427.

Saelens, B. E., & Epstein, L. H. (1996). Reinforcing value of food in obese and non-obese women. *Appetite, 27,* 41-50.

Shigemura, N., Ohta, R., Kusakabe, Y., Miura, H., Hino, A., Koyano, K., Nakashima, K., & Ninomiya, Y. (2004). Leptin modulates behavioral responses to sweet substances by influencing peripheral taste structures. *Endocrinology, 145,* 839-847.

Shimizu, H., Shimomura, Y., Nakanishi, Y., Futawatari, T., Ohtani, K., Sato, N., & Mori, M. (1997). Estrogen increases in vivo leptin production in rats and human subjects. *J Endocrinol, 154,* 285-292.

Smith, J. A., & Epstein, L. H. (1991). Behavioral economic analysis of food choice in obese children. *Appetite, 17,* 91-95.

Solomon, S. J., Kurzer, M. S., & Calloway, D. H. (1982). Menstrual cycle and basal metabolic rate in women. *Am J Clin Nutr, 36,* 611-616.

Stunkard, A. J., & Messick, S. (1985). The three-factor eating questionnaire to measure dietary restraint, disinhibition and hunger. *J Psychosom Res, 29,* 71-83.

Tarasuk, V., & Beaton, G. H. (1991). Menstrual-cycle patterns in energy and macronutrient intake. *Am J Clin Nutr, 53,* 442-447.

Teirmaa, T., Luukkaa, V., Rouru, J., Koulu, M., & Huupponen, R. (1998). Correlation between circulating leptin and luteinizing hormone during the menstrual cycle in normal-weight women. *Eur J Endocrinol, 139,* 190-194.

Thong, F. S., McLean, C., & Graham, T. E. (2000). Plasma leptin in female athletes: relationship with body fat, reproductive, nutritional, and endocrine factors. *J Appl Physiol, 88,* 2037-2044.

Torrubia, R., Avila, C., Molto, J., & Caseras, X. (2001). The sensitivity to punishment and sensitivity to reward questionnaire (SPSRQ) as a measure of Gray's anxiety and impulsivity dimensions. *Personality and Individual Differences, 31,* 837–862.

Tucci, S. A., Murphy, L. E., Boyland, E. J., Dye, L., & Halford, J. C. (2010). Oral contraceptive effects on food choice during the follicular and luteal phases of the menstrual cycle. A laboratory based study. *Appetite, 55,* 388-392.

Uher, R., Treasure, J., Heining, M., Brammer, M. J., & Campbell, I. C. (2006). Cerebral processing of food-related stimuli: effects of fasting and gender. *Behav Brain Res, 169,* 111-119.

Van Vugt, D. A. (2009). Brain imaging studies of appetite in the context of obesity and the menstrual cycle. *Hum Reprod Update, 16,* 276-292.

Wang, G. J., Volkow, N. D., Logan, J., Pappas, N. R., Wong, C. T., Zhu, W., Netusil, N., & Fowler, J. S. (2001). Brain dopamine and obesity. *Lancet, 357,* 354-357.

Webb, P. (1986). 24-hour energy expenditure and the menstrual cycle. *Am J Clin Nutr, 44,* 614-619.

Weingarten, H. P. (1985). Stimulus control of eating: implications for a two-factor theory of hunger. *Appetite, 6,* 387-401.

Welt, C. K., Chan, J. L., Bullen, J., Murphy, R., Smith, P., DePaoli, A. M., Karalis, A., & Mantzoros, C. S. (2004). Recombinant human leptin in women with hypothalamic amenorrhea. *N Engl J Med, 351,* 987-997.

Wurtman, J. J., Brzezinski, A., Wurtman, R. J., & Laferrere, B. (1989). Effect of nutrient intake on premenstrual depression. *Am J Obstet Gynecol, 161,* 1228-1234.

Wurtman, R. J., & Wurtman, J. J. (1989). Carbohydrates and depression. *Sci Am, 260,* 68-75.

Zheng, H., & Berthoud, H. R. (2007). Eating for pleasure or calories. *Curr Opin Pharmacol, 7,* 607-612.

APPENDIX 1:

PARTICIPANT CONSENT FORM

Université d'Ottawa · University of Ottawa
Faculté des sciences de la santé
École des sciences de l'activité physique
Faculty of Health Sciences
School of Human Kinetics

CONSENT FORM

THE EFFECTS OF MENSTRUAL CYCLE DETERMINANTS ON ENERGY BALANCE IN WOMEN WITH DIFFERENT ADIPOSITY LEVELS

Principal Investigator: Jessica McNeil, M.Sc. (candidate)
Supervisor: Éric Doucet (Ph.D.)
Faculty of Health Sciences, University of Ottawa
School of Human Kinetics

1. INVITATION TO PARTICIPATE: You are invited to participate in the above named research study conducted by Éric Doucet Ph.D, and Jessica McNeil M.Sc. (candidate).

2. PURPOSE OF THE STUDY: The primary purpose of this study is to evaluate the possible effects of a high body adiposity level, as well as an adiposity level within the normal recommended range, on the variations in energy intake and energy expenditure across the different phases of the menstrual cycle. The present study will also explore certain secondary factors which may be related to these variations in energy balance, such as variations in female reproductive hormones, food reinforcement, certain olfactory (smell) factors and leptin levels across the menstrual cycle.

3. BACKGROUND: Most of the studies evaluating appetite and energy variations across the menstrual cycle have focused on women with a body mass index (BMI) within the normal, recommended range. This being said, no studies to date have evaluated the variations in energy balance across the menstrual cycle in overweight and obese women, while only one study compared female reproductive hormones and leptin levels in both lean and obese women. Point in fact; it has been previously demonstrated that there are clear variations in energy intake across the different phases of the menstrual cycle in lean women. However, no significant changes in body weight and fat percentage across the menstrual cycle have been noted in this same population. This being said, there may be different or even more pronounced variations in energy intake in women with high adiposity levels, which may ultimately affect cyclic body weight and/or fat percentage in this population. Measurements during this protocol will include body composition, energy intake, resting energy expenditure, total energy expenditure, testing related to olfactory capacity, food reinforcement, female reproductive hormone and leptin levels, as well as certain psychological and behavioral assessments. Results obtained from this study will enable us to better understand the physiological and behavioral changes that occur across the different phases of the menstrual cycle in conjunction with different adiposity levels, and will help us determine whether women with a high adiposity level may present different or even more pronounced variations in energy intake. Moreover, the evaluation of certain

secondary factors, such as the variations in female reproductive hormones, the occurrence and severity of the premenstrual syndrome (PMS), dietary restraint levels, food reinforcement, olfactory (smell) capacity, and leptin levels may help us elucidate why these variations in energy balance occur across the menstrual cycle. Finally, the findings of this study may have a good practical significance, by providing useful information when building an adequate weight management program for pre-menopausal women, which not only takes into account certain physiological mechanisms related to feeding and the menstrual cycle, but which also tailors to an overweight/obese population.

4. DESCRIPTION OF THE STUDY: Initial visit: During this time, you will be informed of the experimental procedures which will be employed during each experimental session, as well as the equipment which will be used in order to obtain the necessary data. The consent form will then be explained to you, and you may also choose to bring the consent form home so further reading and discussion with family members is made possible. Other questionnaires will also have to be filled out for screening purposes and your height, weight and body composition (Dual Energy X-ray Absorptiometry) will also be measured. Results from these measurements will then be analyzed in order to determine if you correspond to the inclusion criteria of this present study. If you do correspond and agree to participate in this study, you will be asked to come to the research unit for 3 identical experimental sessions over one complete menstrual cycle, with each session lasting approximately 10 hours. Each session will also be held during a different phase of the menstrual cycle: once during menstruation, once prior to ovulation and once during the mid-luteal phase. The procedure which will be employed during each experimental session of testing is described in detail below.

Experimental session

A. Arrival at the laboratory (8:00am-8:15am)—You will arrive at the laboratory from an overnight fast from 8:00pm the previous evening. Your body weight and body composition (Dual Energy X-ray Absorptiometry) will be recorded and a single fasting blood sample will be taken by a qualified nurse in a sterilized environment. In addition, your basal temperature will be measured orally through the use of a digital thermometer.

B. Resting Energy Expenditure (8:15am-9:00am)—After a 20 minute resting period in the supine position, a measurement of resting energy expenditure will be done. The measurement of resting metabolic rate takes place early in the morning after an overnight fast. A plexiglass hood will be placed over your head. The expired air will be sampled for analysis and percentages of oxygen and carbon dioxide determined for 30 minutes. By measuring the flow rate, we will be able to determine the amount of oxygen that is consumed and derive resting energy expenditure. This test requires that you lie quietly and relaxed in bed for around 30 minutes. There are no risks associated with this procedure.

C. Completion of the Shortened premenstrual assessment form (9:00am-9:15am)—The occurrence and severity of the premenstrual syndrome (PMS) felt during

each phase of the menstrual cycle will be evaluated through the use of a short questionnaire. This questionnaire is used to classify the subjective changes in mood and physical conditions, based on a 6 point visual analogue scale (1 = no change and 6 = extreme change) seen or felt by you at that moment.

D. Completion of the 3 factor eating questionnaire (9:15am-9:30am)—This questionnaire is used in order to determine one's dietary restraint level and evaluates 3 aspects of eating behaviour known as cognitive restraint, uncontrolled eating and emotional eating.

E. Computer Game (i) (9:30am-9:45am)—You will be required to complete a computer task to assess the amount of work done for a particular reinforcer (snack or fruit/vegetable). The computer task is as follows. You will sit in front of a laptop which will present to you 2 slot machine type games: one is for your favorite snack food, and the other is for your favorite fruit or vegetable. You will the get the opportunity to work for food points for either the snack food or fruit/vegetable. To earn the points you must work for them by pressing a button on the mouse, which in turn starts the slot machine. Points are earned when all three objects on the screen match.

F. Computer Game (ii) (9:45am-10:15am)—You will be required to complete a computer task to assess food selection and subjective ratings of various food items. The computer task is as follows. You will sit in front of a laptop with a mouse and keyboard. A series of food pictures will be presented to you and you will be required to answer a series of questions asked by the computer program regarding these foods. All answers are recorded by selecting and clicking with either the mouse or the keyboard.

G. Meal consumption (10:15am-5:30pm)—You will then go into an assigned room for the day. In this room, there is a television, a couple of chairs and a desk. You will be allowed to do any activities (read, watch a movie/DVD, use a computer, ect.) that you may desire, except for any form of training or structured voluntary exercise. Once settled in this room, you will be handed a copy of the food menu which will include a wide variety of foods, from which you will be able to choose the type of food from the menu that you may want to consume. At this time, the food will be prepared by the principal investigator and served to you. You will then have a half hour to consume whatever you may want (out of the food that you have chosen). Also, a food menu will be presented to you at every hour. This being said, if there is a food item that you did not get to eat or choose during the last session, then you can always request it in the next meal consumption session.

H. Smell tests (2:00pm-3:00pm)—You will be required to complete 3 different odour tests, which include an odour threshold test, an odour discrimination test and an odour identification test. For the odour threshold test, a set of 3 capsules will be subsequently presented to you and you must identify which of the 3 capsules presents an odour (the other 2 capsules are odourless). As for the odour discrimination test, a set of 3 capsules will be subsequently presented to you and you must identify which of the 3 capsules presents a different odour (the other 2 capsules will have the same odour).

Finally, for the odour identification test, you will have a booklet with multiple choice answers on each page (one page corresponds with one odour capsule). You will be required to identify the correct odour released by the capsule, based on the multiple choice answers that will be provided to you. Examples of some of the odours which will be presented to you are rose, marker, banana, and grass.

I. Food journal and accelerometer (5:30pm-5:45pm)—At the end of each experimental session, a 3-day food journal and an accelerometer will be given to you in order to note food intake and voluntary energy expenditure respectively outside of the laboratory sessions. You will be instructed on how to properly use each tool, and you will be asked to bring back the food journal and the accelerometer at your next experimental session.

J. End of Session 5:45pm.

5. POSSIBLE RISKS/DISCOMFORTS:

The risks associated with this project are low and minimal. The measure of body composition presents a low risk for you. However, it is important to underline that this apparatus will expose you to a minimal radiation (the equivalent of a day in the sun – 0.02 – 0.05 millirem). The blood samples also present very few risks. However, a small local hematoma (a bruise at the venal puncture) could develop during the few days following the blood sampling. It is important to note that the risks of infection, of phlebitis (inflammation of the vein) and vaso vagal shock (loss of consciousness) are very low, but still remain a possibility. There is also no risk associated with the basal metabolic rate measurement, the wearing of an accelerometer, the measurement of basal temperature orally using a digital thermometer, as well as the smelling of the odors which will be presented to you during each of the smell tests.

6. BENEFITS:

Your participation in this study will allow you to gather information regarding the possible variations in energy intake and energy expenditure across the different phases of your menstrual cycle. In addition, the evaluation of certain secondary factors, such as the variations in female reproductive hormones, the occurrence and severity of the premenstrual syndrome (PMS), food reinforcement, olfactory (smell) capacity, and leptin levels may help explain why certain variations in energy balance may occur. Moreover, the results concerning the variations in total and resting energy expenditure, as well as energy intake across the menstrual cycle could help you in tailoring a personal weight management or exercise program which takes into account certain physiological mechanisms related to feeding and the menstrual cycle.

7. MONETARY COMPENSATION:

Parking at the research center is free for participants, as are all scientific tests. In addition, you will be compensated $100.00 for your participation in this study which will be prorated for the number of sessions that are completed. You will not be compensated for missed sessions.

8. CONFIDENTIALITY AND ANONYMITY:

In order to guarantee the confidentiality and anonymity of participants, all precautions and necessary measures will be taken to ensure that results and personal information of participants is kept under the strictest of confidentiality.

-Only the following persons will have access to the material: Supervisor and Principal investigator. Any other individuals involved in the study will not have access to participant's personal information and results.

-The names of participants will not appear on any reports. A number code will be used to identify participants on all research documents.

-All material and information which can be linked to participants will not be made public and will be kept under the strictest confidentiality.

-Participants will not be identified in any way in publications or reports.

-The data collected will be kept in a locked cabinet in the Behavioral and Metabolic Research Unit with restricted access where all participant's folders will be kept. In addition, the computer files will be protected by a password.

-Data will be destroyed five years after publication of study results.

9. VOLUNTARY PARTICIPATION

You are free to refuse to participate and if you choose to participate, you are free to withdraw from the study at any time for any reason. At any moment during this study, the best interests of participants will always prevail upon the objectives of the study. The participants will be made aware of new findings that might influence their decision to take part in the present study.

Any information about your rights as a research participant may be addressed to: Protocol officer for ethics in research, University of Ottawa, 550 Cumberland, Tabaret Hall, room 159, Ottawa, Ontario, K1N 6N5; Phone: (613) 562-5841, email: ethics@uottawa.ca.

If I have any questions about the conduct of the research project, I may contact the research coordinator Jessica McNeil, jmcne097@uottawa.ca, at 613-237-1479.

There are two copies of the consent form, one of which I may keep.

Please choose one of the following options:

If I choose to withdraw from the study, I want that all data gathered from me until the time of withdrawal be destroyed ☐

Even if I withdraw from the study, I accept that the data gathered from me be used for this study ☐

SUPERVISOR'S SIGNATURE
Eric Doucet, Ph.D.: _____ Date: _____

PRINCIPAL INVESTIGATOR'S SIGNATURE

Jessica McNeil, M.Sc. (candidate): _____ Date: _____

PARTICIPANT'S SIGNATURE:
I agree to participate in this study,

_____ _____ Date:_____
Printed Name Signature

APPENDIX 2:

FOOD MENU AND MACRONUTRIENT BREAKDOWN

☐ Croissant
☐ Nature Bagel
☐ Whole wheat bagel with sesame seed
☐ White bread
☐ Whole wheat bread
☐ Packaged oats

☐ Orange
☐ Apple
☐ Banana
☐ Green grapes

☐ Raisin Bran
☐ Corn Flakes
☐ Harvest crunch cereal
☐ Honey nuts cheerios

☐ Valley nature sweet and salty granola bar (grilled mixed nuts)
☐ Chocolate chip dipps (Quaker)
☐ Nutri-grain blueberry bar

☐ Tropicana apple juice
☐ Tropicana orange juice

☐ Pepsi
☐ 7up

☐ Vanilla ice cream
☐ Chocolate ice cream

☐ Skittle
☐ Kit Kat
☐ Caramilk
☐ Hershey chocolate with almonds
☐ 70% Black chocolate
☐ Chocolate chip cookies

☐ Lays nature chips
☐ Lays BBQ chips

☐ Water
☐ 1% milk
☐ 3.25% milk
☐ Chocolate milk
☐ Butter
☐ Silhouette 0% yogurt
☐ Yogurt Danone

☐ Red pepper
☐ Baby carrots
☐ Cucumber
☐ Dip for vegetables

☐ Cheddar cheese
☐ Brie cheese

☐ Breton original crackers

☐ 3 cheese pizza
☐ Meat lasagna
☐ Marinara grilled chicken
☐ Sweet sesame chicken
☐ Chicken pot pie
☐ Beef pot roast

☐ Vegetable soup
☐ Chicken noodle soup
☐ Beef and vegetable soup

☐ Creamy peanut butter
☐ Cream cheese
☐ Strawberry jam

☐ Salt
☐ Pepper
☐ Mustard
☐ Mayonnaise
☐ Ketchup

Food Item	Energy (Kcal/kg)	Protein (g/kg)	%	Dietary fat (g/kg)	%	Carbohydrate (g/kg)	%
Croissant	3521.13	70.42	8.0	183.10	47.0	394.37	45.0
White bagel	2666.67	88.89	13.4	27.78	9.4	511.11	77.1
Whole wheat bagel with sesame	2555.56	88.89	13.8	38.89	13.6	466.67	72.6
seed	2500.00	83.33	13.2	27.78	9.9	486.11	76.9
White bread	2564.10	115.38	18.5	32.05	11.6	435.90	69.9
Whole wheat bread	661.85	24.07	14.5	12.03	16.4	114.32	69.1
Packaged oats**	460.00	10.30	8	0.90	1.6	116.30	90.4
Orange	920.00	10.30	4	4.80	4.2	234.30	91.7
Banana	590.00	1.90	1.2	3.60	5	152.50	93.8
Apple	690.00	7.20	3.8	1.60	1.9	181.00	94.4
Green grapes	3220.34	101.69	10.6	25.42	6	796.61	83.4
Honey nut All bran cereal	3666.67	66.67	7.1	0.00	0	866.67	92.9
Corn Flakes	4888.89	88.89	7.2	200.00	36.7	688.89	56.1
Harvest crunch cereal	3793.10	68.97	7.3	34.48	8.3	793.10	84.4
Honey nut cheerios	4857.14	85.71	7.1	228.57	42.9	600.00	50
Valley nature sweet and salty	4516.13	64.52	5.5	161.29	31	741.94	63.4
granola bar	3513.51	----	0	81.08	22	648.65	78
" Chewy Quaker" chocolate	480.00	4.00	3.3	----	0	116.00	96.7
granola bar	440.00	8.00	6.9	----	0	108.00	93.1
Nutri-grain blueberry bar	440.00	----	0	----	0	116.00	100
Tropicana apple juice	450.70	----	0	----	0	121.13	100
Tropicana orange juice	2240.00	40.00	7.2	152.00	61.3	176.00	31.5
Pepsi	2160.00	40.00	7.3	144.00	59.1	184.00	33.6
7 up	4000.00	2.50	0.3	37.50	8.5	900.00	91.2
Vanilla ice cream**	5111.11	66.67	5.1	266.67	45.8	644.44	49.2
Chocolate ice cream**	4615.38	57.69	4.9	211.54	40.1	653.85	55.1
Skittles	5581.40	116.28	8.2	348.84	55.6	511.63	36.2
Kit Kat	6000.00	60.00	3.8	480.00	69.2	420.00	26.9
Caramilk	4857.14	57.14	4.7	228.57	41.9	657.14	53.5
Hershey chocolate with almonds	5600.00	60.00	4.3	360.00	58.3	520.00	37.4
70% dark chocolate	5200.00	60.00	4.6	300.00	51.3	580.00	44.1
Chocolate chip cookies	----	----	0	----	0	----	0
Lays regular chips	400.00	36.00	33.8	10.00	21.1	48.00	45.1
Lays BBQ chips	640.00	36.00	23.1	32.00	46.2	48.00	30.8
Water	640.00	28.00	17.7	10.00	14.2	108.00	68.1
1% milk	7000.00	----	0	800.00	100	----	0
3.25% milk	350.00	30.00	33.3	----	0	60.00	66.7
1% chocolate milk	900.00	40.00	17.1	15.00	14.4	160.00	68.4
Butter	270.00	8.90	11.5	1.90	5.5	64.30	83
Silhouette 0% yogurt	120.00	5.70	16.6	1.60	10.5	25.00	72.9
Danone 1.5% yogurt	352.94	11.76	12.5	----	0	82.35	87.5
Red pepper	4666.67	33.33	2.9	466.67	91.3	66.67	5.8
Cucumber	4000.00	233.33	23.7	333.33	76.3	----	0
Baby carrots	3000.00	200.00	27.6	233.33	72.4	----	0
Ranch vegetable dip	5000.00	90.91	7.3	227.27	41.3	636.36	51.4
Cheddar cheese	2746.48	98.59	14.5	133.80	44.2	281.69	41.3
Brie cheese	1118.88	83.92	29.6	27.97	22.2	136.36	48.1
Breton original crackers	880.28	73.94	32.8	14.08	14.1	119.72	53.1
3 cheese pizza	1130.14	58.22	20.9	17.12	13.8	181.51	65.2
Meat lasagna	2332.16	67.14	11.6	130.74	50.7	219.08	37.7
Marinara grilled chicken	905.17	47.41	21.5	21.55	22	125.00	56.6
Sweet sesame chicken	400.00	16.00	16.7	----	0	80.00	83.3
Chicken pot pie	400.00	28.00	28.6	8.00	18.4	52.00	53.1
Beef pot roast	520.00	32.00	24	6.00	10.1	88.00	65.9
Vegetable soup	6000.00	200.00	12	533.33	72	266.67	16
Chicken noodle soup	3000.00	66.67	9.1	266.67	81.8	66.67	9.1
Beef and vegetable soup	4000.00	----	0	----	0	933.33	100
Creamy peanut butter	----	----	0	----	0	----	0
Cream cheese	2550.00	109.48	13.2	32.60	8.8	648.09	78
Strawberry jam	660	39.50	21.1	31.10	37.4	77.80	41.5
Salt	7142.86	----	0	785.71	100	----	0
Pepper	1000.00	17.40	6.1	4.90	3.9	257.80	90.1
Mustard							
Mayonnaise							
Ketchup							

APPENDIX 3:

SENSITIVITY TO REWARD QUESTIONNAIRE

Questionnaire: Sensitivity to Punishment and Sensitivity to Reward (SPSRQ)

1. Does the good prospect of obtaining money motivate you strongly to do some things?

   YES NO
2. Do you prefer not to ask for something when you are not sure you will obtain it?
   YES NO
3. Are you often afraid of new or unexpected situations?
   YES NO
4. Is it difficult for you to telephone someone you do not know?
   YES NO
5. Do you often do things to be praised?
   YES NO
6. Do you like being the center of attention at a party or social meeting?
   YES NO
7. In tasks that you are not prepared for, do you attach great importance to the possibility of failure?
   YES NO
8. Do you spend a lot of your time on obtaining a good image?
   YES NO
9. Are you easily discouraged in difficult situations?
   YES NO
10. Are you a shy person?
    YES NO

11. When you are in a group, do you try to make your opinions the most intelligent or the funniest?
    YES NO
12. Whenever possible, do you avoid demonstrating your skills for fear of being embarrassed?
    YES NO
13. Do you often take the opportunity to pick up people you find attractive?
    YES NO
14. When you are with a group, do you have difficulties selecting a good topic to talk about?
    YES NO
15. As a child, did you do a lot of things to get people's approval?
    YES NO
16. Does the possibility of social advancement, move you to action, even if this involves not playing fair?
    YES NO
17. Do you think a lot before complaining in a restaurant if your meal is not well prepared?
    YES NO
18. Do you generally give your preference to those activities that imply an immediate gain?
    YES NO
19. Do you often have trouble resisting the temptation of forbidden things?
    YES NO
20. Whenever you can, do you avoid going to unknown places?
    YES NO
21. Do you like to compete and do everything you can to win?
    YES NO
22. Are you often worried by things that you said or did?
    YES NO
23. Would it be difficult for you to ask your boss for a raise (salary increase)?
    YES NO

24. Do you generally avoid speaking in public?
    YES NO
25. Do you, on a regular basis, think that you could do more things if it was not for your insecurity or fear?
    YES NO
26. Do you sometimes do things for quick gains?
    YES NO
27. Comparing yourself to people you know, are you afraid of many things?
    YES NO
28. Does your attention easily stray from your work in the presence of an attractive stranger?
    YES NO
29. Do you often find yourself worrying about things to the extent that performance in intellectual abilities is impaired?
    YES NO
30. Are you interested in money to the point of being able to do risky jobs?
    YES NO
31. Do you often refrain from doing something you like in order not to be rejected or disapproved of by others?
    YES NO
32. Do you like to put competitive ingredients in all of your activities?
    YES NO
33. Would you like to be a socially powerful person?
    YES NO
34. Do you often refrain from doing something because of your fear of being embarrassed?
    YES NO
35. Do you like displaying your physical abilities even though this may involve danger?

    YES NO

APPENDIX 4:

THREE-FACTOR EATING QUESTIONNAIRE

FOOD HABITS QUESTIONNAIRE

(Stunkard et Messick, 1984)

This questionnaire contains a certain number of propositions.

If you agree with the statement or if you feel like it can be applied to you, check the case TRUE who correspond to the statement.

If you disagree with the statement or if you feel like it does not applied to you, check the FALSE case who correspond to the statement.

You have the choice to answer (or not) certain questions.

		TRUE	**FALSE**
1.	**When I smell a sizzling steak or see a juicy piece of meat, I find it difficult to keep from eating, even if I have just finished a meal.**	o	o
2.	**I usually eat too much at social occasions, like parties and picnics.**	o	o
3.	**I am actually so hungry that I eat more than 3 times per day.**	o	o
4.	**When I have eaten my quota of calories, I am usually good about not eating any more.**	o	o
5.	**Dieting is so hard for me because I just get too hungry.**	o	o
6.	**I deliberately take small helpings as a means of controlling my weight.**	o	o
7.	**Sometimes things just taste so good that I keep on eating even when I am no longer hungry.**	o	o
8.	**Since I am often hungry, I sometimes wish that while I am eating, an expert would tell me that I had enough or that I can have something more to eat.**	o	o
9.	**When I feel anxious, I find myself eating.**	o	o

10. Life is too short to worry about dieting.	o	o
11. Since my weight goes up and down, I have gone on reducing diets more than once.	o	o
12. I often feel so hungry that I just have to eat something.	o	o
13. When I am with someone who is overeating, I usually overeat too.	o	o
14. I have a pretty good idea of the number of calories in common food.	o	o
15. Sometimes when I start eating, I just can't seem to stop.	o	o
16. It is not difficult for me to leave something on my plate.	o	o
17. At certain times of the day, I get hungry because I have gotten used to eating them.	o	o
18. While on a diet, if I eat food that is not allowed, I consciously eat less for a period of time to make up for it.	o	o
19. Being with someone who is eating often makes me hungry enough to eat also.	o	o
20. When I feel "blue", I often overeat.	o	o
21. I enjoy eating too much to spoil it by counting calories or watching my weight.	o	o
22. When I see a real delicacy, I often get so hungry that I have to eat right away.	o	o
23. I often stop eating when I am not really full as a conscious means of limiting the amount that I eat.	o	o
24. I get so hungry that my stomach often seems like a bottomless pit.	o	o
25. My weight has hardly changed at all in the last 10 years.	o	o
26. I am always hungry so it is hard for me to stop eating	o	o

before I finish the food on my plate.

27.	**When I feel lonely, I console myself by eating.**	o	o
28.	**I consciously hold back at meals in order not to gain weight.**	o	o
29.	**I sometimes get very hungry late in the evening or at night.**	o	o
30.	**I eat anything I want, anytime I want.**	o	o
31.	**Without even thinking about it, I take a long time to eat.**	o	o
32.	**I count calories as a conscious means of controlling weight.**	o	o
33.	**I do not eat some foods because they make me fat.**	o	o
34.	**I am always hungry enough to eat at any time.**	o	o
35.	**I pay a great deal of attention to changes in my figure.**	o	o
36.	**While on a diet, if I eat a food that is not allowed, I often then splurge and eat other high calorie foods.**	o	o

PART 2

Please answer the following questions by circling the number that best corresponds to you.

37. How often are you dieting in a conscious effort to control your weight ?

Rarely	Sometimes	Usually	Always
1	2	3	4

38. Would a weight fluctuation of 5lbs (2 kgs) affect the way you live your life ?

Not at all	Slightly	Moderately	Very much
1	2	3	4

39. How often do you feel hungry ?

Only	Sometimes	Often	Almost
At mealtimes	between meals	between meals	always
1	2	3	4

40. Do your feelings of guilt about overeating help you control your food intake ?

Never	Rarely	Often	Always
1	2	3	4

41. How difficult would it be for you to stop eating halfway through dinner and not eat for the next 4 hours ?

Easy	Slightly Difficult	Moderately Difficult	Very Difficult
1	2	3	4

42. How conscious are you of what you are eating ?

Not at all	Slightly	Moderately	Extremely
1	2	3	4

43. How frequently do you avoid « stocking up » on tempting foods ?

Almost Never	Seldom	Usually	Almost always
1	2	3	4

44. How likely are you to shop for low calorie foods ?

Unlikely	Slightly Unlikely	Moderately likely	Very likely
1	2	3	4

45. Do you eat sensibly in front of others and splurge alone ?

Never	Rarely	Often	Always
1	2	3	4

46. How likely are you to consciously eat slowly in order to cut down on how much you eat ?

Unlikely	Slightly Unlikely	Moderately likely	Very likely

1	2	3	4

47. How frequently do you skip dessert because you are no longer hungry ?

Almost Never	Seldom	At least once per week	Almost every day
1	2	3	4

48. How likely are you to consciously eat less than you want ?

Unlikely	Slightly Unlikely	Moderately likely	Very likely
1	2	3	4

49. Do you go on eating binges though you are not hungry ?

Never	Rarely	Sometimes	At least Once per week
1	2	3	4

50. On a scale of 1 to 5, where :

- 0 (zero) means no restraint in eating (eating whatever you want, whenever you want it) and,

- 5 means total restraint (constantly limiting food intake and never "giving in"),

What number would you give yourself?

- **Eat whatever you want, whenever you want it**
  0

- **Usually eat whatever you want, whenever you want it**
  1

- **Often eat whatever you want, whenever you want it**
  2

- **Often limit food intake, but often "give in"**
  3

- **Usually limit food intake, rarely "give in"**
  4

- **Constantly limiting food intake, never "giving in"**
  5

51. To what extent does this statement describe your eating behaviour?

"I start dieting in the morning, but because of many different things that happen during the day, by evening I have given up and eat what I want, promising myself to start dieting again tomorrow"

Not like Me	Little like me	Pretty good description of me	Describes me perfectly
1	2	3	4

APPENDIX 5:

SHORTENED PREMENSTRUAL ASSESSMENT FORM

Shortened Premenstrual Assessment Form

Name: _____ Date: _____

For each of the symptoms below, circle the number that most closely describes the intensity of your premenstrual symptoms. These are symptoms that would occur during the premenstrual phase of your cycle. Rate each item on this list on a scale from 1 (not present or no change from usual) to 6 (extreme change, perhaps noticeable even to casual acquaintances).

	1=No change					Extreme change=6
1. Pain, tenderness, enlargement or swelling of breasts	1	2	3	4	5	6
2. Feeling unable to cope or overwhelmed by ordinary demands	1	2	3	4	5	6
3. Feeling under stress	1	2	3	4	5	6
4. Outburst of irritability or bad temper	1	2	3	4	5	6
5. Feeling sad or blue	1	2	3	4	5	6
6. Backaches, joint and muscle pain, or joint stiffness	1	2	3	4	5	6
7. Weight gain	1	2	3	4	5	6
8. Relatively steady abdominal heaviness, discomfort or pain	1	2	3	4	5	6
9. Edema, swelling, puffiness, or water retention	1	2	3	4	5	6
10. Feeling bloated	1	2	3	4	5	6

Total Score ____