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Energy intake, resting energy expenditure, activity levels, energy balance and serum hormone concentrations in sedentary and athletic males.

By

Kelly Mackenzie 

A thesis submitted to the Faculty of Graduate Studies and Research in partial fulfillment of the requirements for the degree of Master of Science.

Faculty of Physical Education and Recreation

Edmonton, Alberta

Spring 2000

University of Alberta

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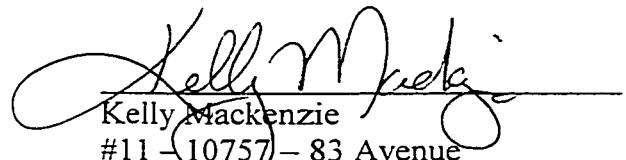

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The undersigned certify that they have read, and recommended to the Faculty of Graduate Studies and Research for acceptance, a thesis entitled **Energy intake, resting energy expenditure, activity levels, energy balance and serum hormone concentrations in sedentary and athletic males** submitted by **Kelly Mackenzie** in partial fulfillment of the requirements for the degree of Master of Science.

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Dated: January 28, 2000

ABSTRACT

Energy intake, resting energy expenditure, activity levels, energy balance and serum hormone concentrations in sedentary and athletic males.

Resting reproductive (total testosterone, free testosterone, LH, FSH, prolactin) and metabolic (T_3, T_4, cortisol) hormone concentrations were assessed in 16 sedentary males (SC, less than 4 hours aerobic training weekly), 20 moderate-endurance trained males (ME, 5 - 10 hours aerobic training weekly), and 16 high-endurance trained males (HE, greater than 11 hours aerobic training weekly). In addition, various energy balance markers (energy intake, resting energy expenditure (REE) and energy expenditure from exercise) were measured in these same 3 groups. Average energy intake (mean ± SD, kcal/day) was higher in HE (3470 ±523) and ME (3445 ±449) than SC (2709 ±513) ($p < 0.05$). REE was not different between groups. Energy expenditure from exercise (mean ± SD, kcal/day) was higher in HE vs. ME or SC and ME vs. SC (HE = 951 ±245; ME = 695 ±150; SC = 413 ±127) ($p<0.05$). Energy balance (calculated as energy intake - [REE + thermic effect of feeding + exercise energy expenditure]) was not different between groups. With exception of T_3, serum hormone concentrations were similar across groups. T_3 (mean ± SD, ng/dl) was significantly lower in HE (102 ± 79.7) than SC (117.8 ±92.3) ($p<0.05$).

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From the bottom of my heart, I would like to thank my friends, family, and Vicki Harber for the persistent encouragement, support and direction you have all provided me throughout this experience. You were all very necessary and important to me!

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LIST OF SYMBOLS AND ABBREVIATIONS

CNS	Central nervous system
DEBQ-R	Dutch eating behaviour questionnaire - Restraint
EDI	Eating disorder inventory
FSH	Follicle stimulating hormone
GnRH	Gonadotropin releasing hormone
HE	High-endurance athlete
H-P-G axis	Hypothalamic-pituitary-gonadal axis
hr/wk	Hours per week
kcal/kgLBM/day	Kilocalories per kilogram lean body mass per day
kcal/kgFFM/day	Kilocalories per kilogram fat free mass per day
kgFFM	Kilogram fat free mass
km/wk	Kilometers per week
LH	Luteinizing hormone
ME	Moderate-endurance athlete
N.S.	Not significant
REE	Resting energy expenditure
RER	Respiratory exchange ratio
RIA	Radioimmunoassay
rpm	Revolutions per minute
SC	Sedentary control
TEF	Thermic effect of feeding
T_3	Triiodothyronine
T_4	Thyroxin
VO_2	Ventilatory oxygen consumption
VO_2max	Maximal ventilatory oxygen consumption

CHAPTER 1

INTRODUCTION

1. PURPOSE

The body of literature examining reproductive function in male athletes is small. However, there is sufficient information suggesting that endurance exercise may suppress hormones of the male reproductive axis as circulating testosterone levels, both total and free, in male endurance athletes are lower compared to sedentary controls (Arce et al, 1993b; Hackney et al, 1988; Wheeler et al, 1991; Wheeler et al, 1986; Wheeler et al, 1984). Luteinizing hormone, responsible for the production of testosterone, has been shown to be altered in frequency or amplitude by training (MacConnie et al, 1986; McColl et al, 1989), while others have found no change (Wheeler et al. 1984; Wheeler et al, 1991). Increased training has also been associated with changes in sperm characteristics (De Souza et al, 1994a). Although the mechanism underlying these physiological changes in active males is unclear, it has been proposed that a particular volume of exercise, once exceeded, may be responsible for this phenomena ("volume threshold") (De Souza et al. 1994a; De Souza et al, 1997).

Similarly, reproductive disorders and menstrual irregularities are well documented within the female athlete population. More information on females is available regarding variables that may lead to this phenomenon. The literature suggests that increased energy expenditure through exercise (Beitins et al, 1991) as well as decreased energy intake (Loucks, 1996; Loucks et al, 1994a; Williams et al,

1995) contribute to changes of the menstrual cycle. An abrupt change in energy availability, defined as acute differences in energy intake to expenditure, or a longer term energy imbalance, defined as a chronic state of insufficient energy intake for total energy expenditure, may lead to an interruption in menstrual function (Cumming et al, 1994; Loucks et al, 1994b) as a means to conserve energy. Observable clinical consequences such as oligomenorrhea or amenorrhea may present. In contrast, disruption to spermatogenesis may be undetected by males.

The research examining chronic states of energy balance in male athletes is limited. Recent studies have examined the effects of energy expenditure on male hormones related to reproductive function however the current literature has not calculated energy balance in male athletes and its impact on the reproductive axis. A "volume threshold" effect was examined in three groups of males and were distinguished using weekly running distance (km/wk and hr/wk) over the past 12 months (De Souza et al, 1994a). High-mileage runners ran a minimum of 104 km/wk, moderate-mileage runners ran between 40 to 56 km/wk. and sedentary controls performed no more than 1 hour of aerobic exercise per week. Decreased testosterone levels and alterations to spermatogenesis were more abundant in high mileage runners when compared to moderate mileage or controls (De Souza et al, 1994a) suggesting that running more than 100 km/wk may represent a "volume threshold" that if exceeded may lead to reproductive disturbances. De Souza et al (1997) further defined running distances for assessing a threshold effect as 1) High-mileage: greater than 100 km/wk and a minimum of 8 hrs/wk; 2) Moderate-mileage: 40 to 90 km/wk for 2 – 6 hrs/wk. Additional clarification is necessary to determine

total training volume (eg. modality, frequency, duration and intensity). Furthermore, the definitions of volume threshold provided by De Souza et al (1994a, 1997) are difficult to apply to sports other than running.

An increase in volume of training has been associated with a suppression of male hormones along the reproductive axis (De Souza et al, 1994a; De Souza et al, 1997). An increased energetic demand from exercise, perhaps superimposed with inadequate energy compensation, may result in a negative energy balance in highly trained male athletes. The parameters of an athlete's energy balance may be further explored beyond volume of exercise by assessing energy intake and eating attitudes as well as resting energy expenditure. Analysis of these variables may also distinguish differences in energy balance and, or hormones of the reproductive axis in high-endurance athletes from moderate-endurance athletes and sedentary controls.

This study intends to examine the relationship between energy intake, energy expenditure and hormones of the reproductive axis in male athletes. Markers of energy balance including energy intake and eating attitudes, resting energy expenditure, costs of physical exercise and daily active living, will be assessed to provide greater insight into an athlete's states of energy balance. Hypothalamic-pituitary-gonadal axis hormones as well as cortisol and markers for energy deficiency (T_3 and T_4) will be measured. These findings will further our understanding energy balance and the accompanying hormonal profile found between groups of athletes when separated by volume of exercise.

2. SIGNIFICANCE OF THE STUDY

Suppressed concentrations of circulating reproductive hormones including reduced levels of testosterone as well as changes in spermatogenesis have been found in endurance-trained athletic males (De Souza et al, 1994a). Males who expend large amounts of energy yet deprive themselves of sufficient energy replenishment are at risk of developing physiological changes to semen including decreased sperm count, decreased sperm mobility, immature sperm development as well as impaired abilities of ovum penetration. Further, there is some research suggesting that decreased levels of testosterone may postpone the onset of puberty in males as well as have negative effects on bone mineral density (Bennell et al, 1996). Although endurance exercise has not been demonstrated to directly cause male infertility (Arce et al, 1993a), there is potential for a negative energy balance to exist in high-endurance males that may lead to physiologically suppressed hormones of the reproductive axis. Identification of factors associated with blunted gonadal steroid production at increased volumes of training may provide an insight to mechanisms underlying this phenomena as well as an opportunity for future prevention as well as reversibility of suppressed hormones and alterations to sperm in male endurance-trained athletes.

3. HYPOTHESIS

The hypotheses will be:

3.1 Circulating testosterone levels (total and free) will be significantly lower in the high-endurance group (HE) than the moderate-endurance group (ME) or the sedentary control (SC);

3.2 Luteinizing hormone and follicle stimulating hormone will be significantly lower in the HE than the ME or SC; prolactin will be significantly lower in the HE than the ME or SC; cortisol will be significantly higher in the HE than the ME or SC; T_3 and T_4 will be significantly lower in the HE than the ME or SC;

3.3 Resting energy expenditure will be significantly lower in the HE than the ME or SC; eating attitudes will not be significantly different between the three groups; energy expenditure reported as total time of training as well as measured with the TriTrac R3-D™ will be significantly higher in the HE than the ME and SC and significantly in higher in the ME than the SC; energy intake will be not be significantly different between the three groups; and energy balance will be a negative value in HE and a positive value for ME and SC.

4. DELIMITATIONS

Fifty-two healthy males, participating regularly in various durations of weekly aerobic training (cycling), with no history of illness or use of medications that are known to influence hormonal status will participate in a 4-day experiment in which

energy expenditure (resting energy expenditure and physical activity), energy intake and eating attitudes will be measured. Following informed consent and initial screening tests for eligibility, subjects will be placed into 1 of 3 groups based upon self-reported habitual hours of aerobic training per week: 1) High-endurance athletes (HE); 2) Moderate-endurance athletes (ME); and 3) Sedentary controls (SC). A single blood sample will be collected and analyzed for testosterone (free and total), luteinizing hormone (LH), follicle stimulating hormone (FSH), prolactin, cortisol, T_3 and T_4. Subjects will complete diet records for 4 days as well as complete 2 questionnaires on eating attitudes. Subjects will record all physical activity in a logbook for 4 days while also wearing a accelerometer (TriTrac R3-D™ monitor) for the same 4 days other than during sleep or water activities (ie. showers or swimming). The TriTrac R3-D™ monitor measures movement and is a valid and reliable method of calculating energy expenditure from exercise (Welk et al, 1995).

5. LIMITATIONS

The nature of this research has several inherent limitations:

5.1 Not true random selection: The participants will be recruited from various sports teams, recreational facilities, the University of Alberta, and word of mouth. The eligibility criteria may guide selection of volunteers in that those who do not meet established criteria will be prevented from participating in this study.

5.2 Not true random assignment: The subjects will be assigned to 1 of 3 groups, based upon self-reported hours of aerobic training per week.

5.3 Experimenter and equipment error: It is recognized that errors in data collection and data analysis are potential limitations to this study. For example, hydrostatic weighing may have a 2-3 % body fat error due to technical error as well as estimations regarding volume of residual air and gastro-intestinal air. Further, the TriTrac R3-D™ may have limitations in its' sensitivity to intensity of movement while cycling as well as its' inability to be worn when swimming.

5.4 Dietary intake records: Although the use of 4-day dietary records has been chosen, it is recognized that there are inherent limitations with this method. Subject compliance and accuracy of self-reporting may be suspect (Mertz et al, 1991). There is potential for misrepresentation of habitual dietary intakes when individuals are conscious of their eating habits. As a result, the 4-day diet record may not represent chronic eating habits.

5.5 Resting energy expenditure (REE): Oxygen consumption at rest (REE test) is recognized as an estimation of energy expenditure. Subject compliance to fasting for 12 hours as well as adhering to a full 24 hours of rest are noted as potential limitations to the accuracy of the estimation of REE.

5.6 Activity logs & TriTrac R3-D™: Use of the TriTrac R3-D™ as well as the logbook will provide estimations of energy expenditure from exercise. The accuracy of these methods may be limited. Subject compliance to wearing TriTrac R3-D™ as well as varying interpretations of intensity of exercise are also limitations.

5.7 Reproductive status: Although subjects will be asked about their known reproductive status on a questionnaire, it is recognized that the subclinical nature of sperm characteristics will be unknown for most males.

5.8 Single blood samples: Analysis of hormones from a single blood sample may be a limitation. Timing during the day for blood sample has been restricted to between 4:00 pm and 6:00 pm to avoid peak levels of testosterone, present in the mornings. However, the pulsatile nature of LH will not be represented in this blood analysis.

6. DEFINITIONS

6.1 Energy Availability: Energy availability is a calculation of the difference between energy intake and energy expenditure over a short period of time (Loucks et al, 1994b). The resulting available energy sustains the following metabolic activities: cell maintenance, locomotion, thermoregulation, growth, and reproduction. Oxidizable metabolic fuels are also stored in adipose tissue (Wade et al, 1996). If available energy is limited, energy will be utilized to where it is needed to maintain basic life function (cell maintenance, locomotion and thermoregulation). Energy for growth, adipose tissue stores and reproductive function may then be jeopardized (Loucks, 1996; Wade et al, 1996).

6.2 Energy Balance: Energy balance is the difference between total energy intake (from foods) and total energy expenditure (from rest, daily active living, thermic effect of feeding and exercise). It is reflective of a chronic energy

state. If energy intake is sufficient for energy expenditure, energy balance is established. Conversely, negative energy imbalance refers to inadequate energy intake for the expenditure performed. In this study, chronic energy intake will be calculated from the average of a 4-day dietary intake record. In addition, chronic energy expenditure will be determined from an accelerometer used to measure energy expenditure from exercise (averaged over 4 days) as well as measured resting energy expenditure and estimated thermic effect of feeding. Energy balance can thus be determined by subtracting average total daily energy expenditure from average total daily energy intake.

6.3 Volume Threshold: Beyond an identified volume of endurance training, there is an increased tendency for suppresses hormonal profiles of the H_P-G axis as well as alterations to spermatogenesis in males. This volume has been identified in male runners as greater than 104 kilometers per week (De Souza et al. 1994a). These athletes presented with significantly lower testosterone and alterations to spermatogenesis than males running 40 to 56 kilometers per week or sedentary controls.

6.4 High-endurance athletes: Endurance trained males who have self-reported aerobic training of a minimum of $11 - 15^{+}$ hours per week and whose predominant training modality is cycling.

6.5 Moderate-endurance athletes: Endurance trained males who have self-reported aerobic training between 5 – 10 hours per week and whose predominant training modality is cycling.

6.6 Sedentary control males: Untrained, healthy men who have self-reported less than 4 hours per week of aerobic exercise.

CHAPTER 2

REVIEW OF LITERATURE

1. INTRODUCTION

Decrease in gonadal steroid production as well as altered spermatogenesis have been identified in male endurance athletes (Arce et al, 1993b; De Souza et al, 1994a; Hackney et al, 1990; Hackney et al, 1988; Jensen et al, 1995; McColl et al, 1989; Wheeler et al, 1991; Wheeler et al, 1986; Wheeler et al, 1984). Male athletes may be unaware of subclinical alterations to the male reproductive axis as most are not detectable without clinical evaluation (Arce et al, 1993a; Cumming, 1989b; De Souza et al, 1994a).

Insufficient energy intake and excessive volume of training may lead to a negative energy balance and have been identified as potential variables impeding gonadal function in both male and female athletes. As successful reproductive function requires energy (Bisdee et al, 1989; Wade et al, 1996), inadequate energy may lead to physiological problems. Determination of energy balance may further the understanding of relationships existing between energy intake, exercise energy expenditure and the reproductive axis. Currently, volume of training thresholds (De Souza et al, 1994a; De Souza et al , 1997) and energy availability models (Loucks et al, 1994b) have been described and provide insight to the roles of exercise and energy intake on changes in reproductive function. The literature describing the mechanism(s) underlying suppressed hormones of the reproductive axis and the possible role of negative energy balance in male athletes is sparse. The energy

balance literature in women athletes with suppressed reproductive hormone profiles is more abundant, hence these findings may help address this phenomena in males.

A discussion of normal male reproductive physiology, physiological changes to the reproductive system in active males as well as the role of energy balance in the male reproductive axis will follow. The exploration of energy balance will include energy intake and eating attitudes of male athletes. As well, the relationships between total daily energy expenditure including resting energy expenditure, thermic effect of feeding and exercise energy expenditure and suppressed gonadal hormone production in the male endurance athlete will be examined. Some discussion surrounding the physiological adaptations of the reproductive system in females will be presented to provide additional insight to the energetic challenge faced by male athletes.

2. MALE REPRODUCTIVE PHYSIOLOGY

2.1 Normal reproductive physiology

The hypothalamic-pituitary-gonadal axis (H-P-G axis) is responsible for maintenance of normal reproductive function in both males and females. Normal reproductive function in males requires adequate healthy hormonal production and secretion that results in healthy gamete production. A conceptual figure of the interaction of the hormones responsible for the reproductive system in males aids in understanding the roles of each hormone (Figure 1). The hypothalamus secretes gonadotrophin releasing hormone (GnRH) which in turn stimulates the anterior pituitary gland to produce and secrete luteinizing hormone (LH) and follicle-

stimulating hormone (FSH). GnRH and LH are both released in a pulsatile manner. Normal adult pulse frequency of GnRH is approximately every 15 minutes. Larger pulse amplitudes of LH occur approximately every 70 to 96 minutes in response to the release of GnRH (Filicori, 1986). LH stimulates the production of testosterone in the Leydig cells, which are present in the interstitial spaces within the seminiferous tubules of the gonads (Hadley, 1988). LH concentrations have been positively correlated to a lagged time release of testosterone (Veldhuis et al, 1987). Testosterone appears to be secreted in a both a pulsatile and circadian pattern (Veldhuis et al, 1987). Presence of FSH as well as testosterone stimulate spermatogenesis within the Sertoli cells of the seminiferous tubules (Hadley, 1988).

The H-P-G axis in males is regulated by a negative feedback loop. Inhibin, which is secreted by the Sertoli cells in the testes, inhibits FSH production. Testosterone released from the testes can also control the axis by acting at the pituitary and hypothalamus (refer to Figure 1). Prolactin is thought to enhance LH reception at the Leydig cell (Hadley, 1988). Increased cortisol levels may inhibit testosterone production while other adrenal and neural hormones may also contribute to the inhibition or stimulation of production of hormones within the H-P-G axis (Cumming et al, 1983). Amenorrheic runners present with mild hypercortisolism in comparison to eumenorrheic runners or controls, suggesting the adrenal axis may mediate disruption to the H-P-G axis through increased stress and, or decreased adrenal sensitivity (De Souza et al, 1994b; Loucks et al, 1989).

Testosterone is an androgen that is responsible for the development and maintenance of primary and secondary sex characteristics in males. These

characteristics include sexual organ development, hair growth, vocal deepening as well as skeletal development (Hadley, 1988; Mountcastle, 1980). Further, testosterone has effects on the central nervous system including sexual drive, libido as well as physical vigor (Hadley, 1988; Mountcastle, 1980).

Testosterone is a protein-bound steroid hormone. Approximately 50 percent of testosterone is bound to albumin while 45 percent is bound to sex-hormone binding globulin (SHBG) (Goodman, 1994). Free testosterone is unbound and accounts for the remainder of total testosterone. Ninety-five percent of plasma testosterone is produced in the testes while the remainder is produced by the adrenal gland (Mountcastle, 1980). Normal ranges of plasma testosterone in adult males are between 300 – 1000 ng/dl while plasma LH levels are between 5 – 20 mIU/ml (Wilson et al, 1985). Other circulating hormone ranges for healthy males are presented within tables 12 and 13 (Chapter 4).

Sperm, which is manufactured in the seminiferous tubules of the testes, carry and transmit the male genetic material to the female egg for conception. Quality of spermatogenesis is determined by sperm morphology, mobility and penetration capabilities as well as sperm count. These characteristics are important for evaluating fertility. Adequate hormonal regulation provided by the H-P-G axis is essential for spermatogenesis to occur.

2.2 Alterations of male reproductive physiology

Physiological suppression of hormones as well as alterations to sperm often remains undetected (Arce et al, 1993a; Cumming, 1989b; De Souza et al, 1994a).

Disrupted testicular androgenesis and spermatogenesis are not as apparent compared to those symptoms that have been observed with the female reproductive system (Cumming et al, 1994). Interruption to the H-P-G axis may occur at any level. The adrenal axis, the role of prolactin and other neuroendocrine disturbances may also contribute toward the suppression of circulating testosterone. It has been suggested that the reduction in testosterone within certain populations may be explained by a decrease in production of the gonadal steroid or the increase in clearance of the hormone (Cumming et al, 1989b; De Souza et al, 1997; Wheeler et al, 1984). Within the male athletic population, there remains uncertainty as to the mechanisms that lead to the disruption of the H-P-G axis.

Decreased circulating levels of testosterone may affect development and maintenance of secondary sex characteristics. Low androgen levels may also result in decreased sex drive and libido. Bone density has not been surveyed in males to the same extent as females, with respect to suppressed reproductive hormone profiles. However it is possible that reduced levels of circulating testosterone may affect skeletal development and maintenance as well as postpone the onset of puberty in males (Bennell et al, 1996). Sperm quality is also at risk, including altered and, or reduced sperm density, sperm motility, sperm morphology and in vitro penetration. These characteristics, specifically reduction of in vitro penetration capabilities, pose as potential complications associated with infertility (Arce et al, 1993a). Further understanding of the mechanisms responsible for alterations to the H-P-G axis and testicular function is needed.

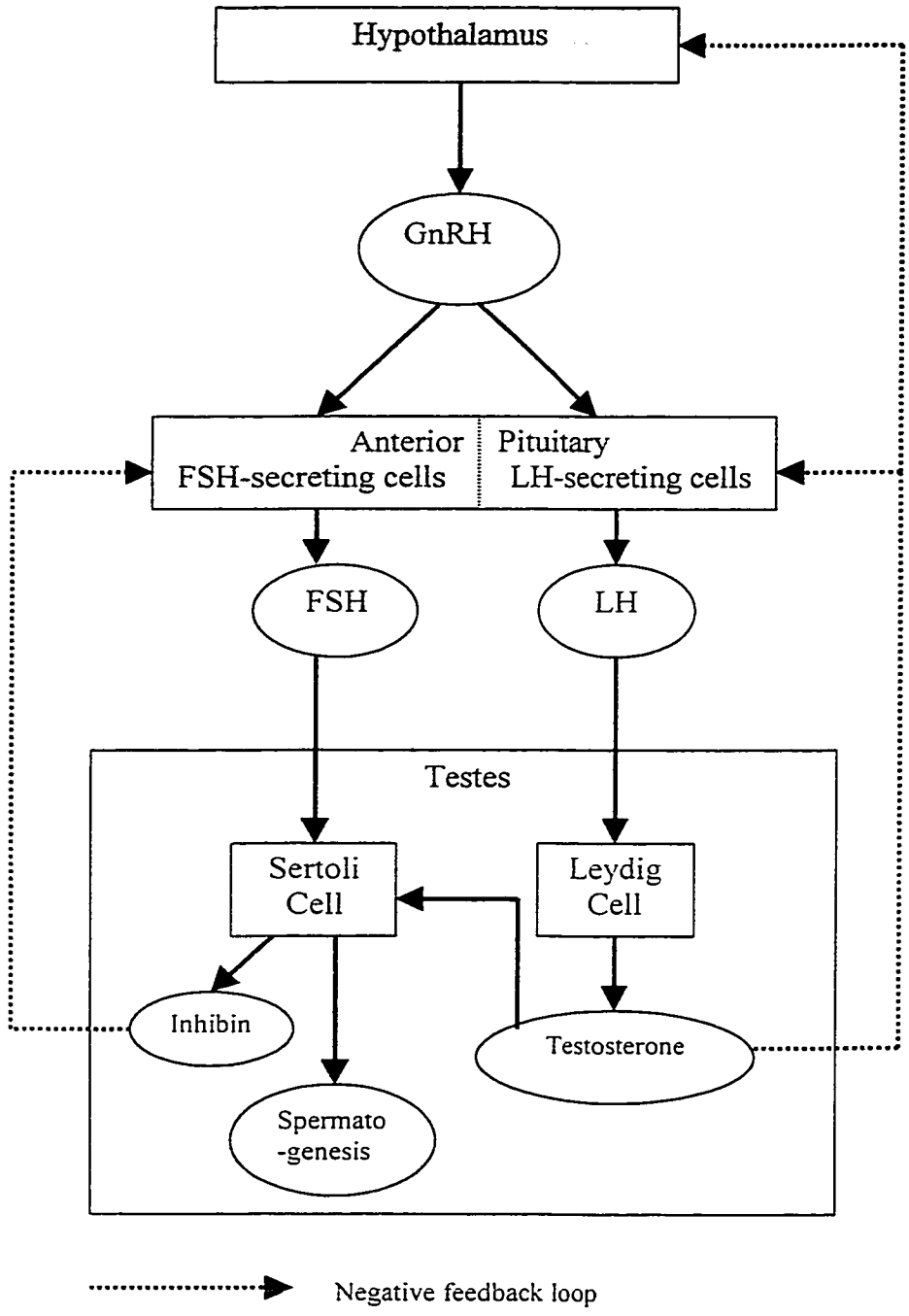


Figure 1. Control of testicular function. (Modified from Sherwood, 1991)

2.3 Alterations of reproductive physiology in male athletes

There is sufficient literature suggesting that male endurance athletes have a greater tendency for physiological changes of the reproductive axis compared to healthy sedentary males. High volumes of aerobic training are correlated with decreased levels of testosterone, altered LH production and release as well as decreased prolactin and increased cortisol (Arce et al, 1993b; De Souza et al, 1994a; Griffith et al, 1990; Hackney et al, 1988; Hackney et al, 1990; Jensen et al, 1995; McColl et al, 1989; Roberts et al, 1993; Wheeler et al, 1984; Wheeler et al, 1991). Further, in comparison to non-athletic males, endurance-trained males have a greater tendency for decreased sperm production, motility and altered sperm morphology (Arce et al, 1993b; De Souza et al, 1994a; Lucia et al, 1996). Similarly, female endurance-trained athletes also express a higher frequency of physiological changes to the reproductive axis when compared to healthy sedentary controls. Table 1 provides a summary of the literature regarding training and specific hormone profiles in male athletes.

Research exploring predisposing factors of alterations to reproductive hormones in male athletes is limited. Potential pathways leading to alterations in male reproduction have been proposed (Figure 2). Figure 2 illustrates the potential locations along the H-P-G axis where disturbances may occur for male athletes as well as illustrates potential outcomes of these disturbances including changes to endocrine profiles as well as to sperm quality (Arce et al, 1993a). Within the female athletic population, there is more information providing insight to alterations to the reproductive system. While one direct mechanism has not been identified, several

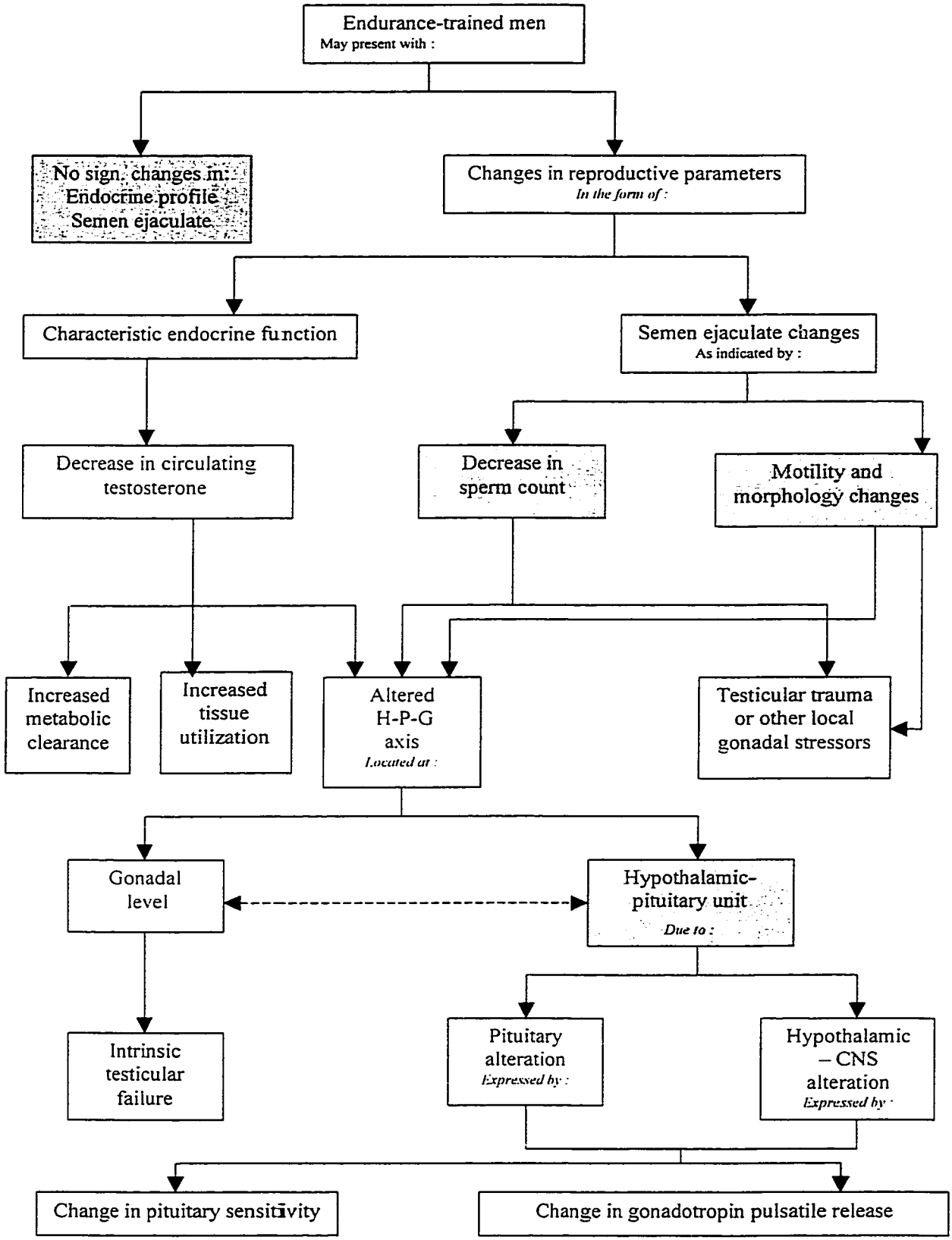


Figure 2. Potential alterations of reproductive function in male athletes. (Modified from Arce et al. 1993a)

Table 1 – Resting hormone concentrations in male endurance-trained athletes compared to sedentary controls.

	Type of Study	Exercise parameters	Total Testosterone	Free Testosterone	LH	FSH	Prolactin	Cortisol
Arce et al, 1993b	Cross-sectional	Trained = 9.7 hrs/wk; 109.2 km/wk	Decreased	Decreased	NS	NS	NS	-
De Souza et al, 1994a	Cross-sectional	Highly trained = >108km/wk (9.7 hrs/wk)	Decreased	Decreased	NS	NS	NS	NS
Griffith et al, 1990	2 week prospective	Training increase = overload total km/wk (overtraining)	Decreased	-	-	-	-	-
Hackney et al, 1988	Cross-sectional	Trained = 68.5 min/day; 6.6 days/wk	Decrease	Decrease	NS	-	NS	NS
Hackney et al, 1990	Cross-sectional	Trained = 17 km/day	Decreased	NS	NS	-	NS	NS
Jensen et al, 1995	Longi-tudinal	Training increase = 60 – 100km/wk	NS	-	NS	NS	NS	-
McColl et al, 1989	Cross-sectional	Trained = > 80 km/wk	Decreased	-	Decreased (area under curve, amplitude)	-	-	-
Roberts et al, 1993	Inter-vention	Training increase = increase total volume (overtraining)	Decreased	-	-	-	-	Increased
Wheeler et al, 1984	Cross-sectional	Trained = > 64 km/wk	Decreased	Decreased	NS	NS	Decreased	NS
Wheeler et al, 1991	Longi-tudinal	Training increase = 56 km/wk from 0 km/wk	Decreased	-	NS	NS	Decreased	-

Note: N.S. - Not significant

predisposing factors have been identified in female athletes experiencing menstrual irregularities. These include low body weight, low body fat, and increased stress levels (both physical and emotional) (Cumming et al, 1994). Other literature has examined the relationship between energy balance, energy intake and energy expenditure with reproductive hormonal profiles and menstrual cycle alterations in females (Beidleman et al, 1995; Horton et al, 1994). Within the male athletic population, research to date has been primarily restricted to volume of training when examining reproductive hormones. Further discussion of energy balance and its markers will follow.

3. VARIABLES AFFECTING REPRODUCTIVE PHYSIOLOGY

High levels of physical activity or stress as well as low body weight in males may affect fertility (Cumming et al, 1994). Where short-duration, intense exercise results in increased serum testosterone, prolonged submaximal exercise suppresses circulating serum testosterone (Cumming, 1989b). Long-term effects of endurance exercise in males often result in continual suppression of serum testosterone (Arce et al, 1993a; Cumming, 1989b). Some female athletes also display blunted gonadal steroid levels (Harber et al, 1998; Loucks et al, 1989; Lund-Hetland et al, 1993). Similar mechanisms may be operating in both males and females, hence data derived from women may facilitate uncovering factors responsible for reproductive alterations in males. In particular, the role of energy balance and its influence on reproductive hormonal profiles in female athletes has received much attention.

Similar investigations in males may elucidate the role of energy balance and reproductive hormonal profiles in males.

There is no literature that has calculated energy balance, encompassing energy intake, resting energy expenditure, thermic effect of feeding and exercise energy expenditure in males. Research has not been able to support that highly trained endurance male athletes are in a chronic state of negative energy balance, nor the potential impacts of this on the male reproductive system. Several studies of female athletes have evaluated energy balance (Beidleman et al, 1995; Horton et al, 1994), however standardized methods have not been established. Energy intake, food sources and eating attitudes of male athletes also require further research. Similarly, accurate measurements of energy expenditure from all sources (at rest, thermic effect of feeding, daily activity and physical activity) would be beneficial in understanding energy balance. Such in-depth exploration is required in the prevention and reversibility of physiological disruptions to the male reproductive system. Literature pertaining to energy balance markers will follow.

3.1 Energy Balance

Negative energy balance (defined as insufficient energy intake to meet the demands of energy expenditure) may be responsible for alterations in the female reproductive system (Beidleman et al, 1995). In absence of sufficient energy to support the menstrual cycle (Bisdee et al, 1989), the body may try to conserve energy through suppressing involvement of the reproductive system (Loucks et al, 1994b; Wade et al, 1996). Further, insufficient energy intake for energy expended has

resulted in no change to body weight (Drinkwater et al, 1984; Myerson et al, 1991), suggesting that energy conservation may be occurring. Others have found no evidence of energy conservation in female athletes (Wilmore et al, 1992). Although data are limited in the male population, information suggests that low energy intake as well as high energy expenditure may result in altered H-P-G endocrine profiles in males (De Souza et al ,1994a; McColl et al, 1989; MacConnie et al, 1986; Rojdmark et al, 1987; Cameron et al, 1991).

Energy availability (defined as dietary energy intake minus energy expenditure during exercise over a short period of time) has been studied in females (Loucks et al, 1994b). Circulating triiodothyronine (T_3) was evaluated after a 4-day energy availability treatment in females (Loucks et al. 1994b). Reduced levels of T_3, a marker of energetic challenge, were observed in subjects who received no dietary compensation for the exercise performed. Energy deficiency may be alleviated in endurance-trained females by increased energy intake. This may prevent the compensatory response of the H-P-G axis to conserve energy expenditure (Loucks et al, 1994b). There are no data at present investigating similar energetic challenges in male athletes.

Energy balance requires an examination of energy intake in addition to energy expenditure. Total energy expenditure encompasses energy expenditure from rest (REE), thermic effect of feeding (TEF) (calculated at 10% of REE), as well as energy from daily activity and physical activity. Negative energy balance, resulting from insufficient energy intake relative to total energy expenditure, may be a catalyst for disrupting the H-P-G axis (Loucks et al, 1994a). Although some aspects of energy

balance have been evaluated, no study to date has presented a complete analysis of energy balance in male athletes and related that to reproductive hormone profiles. In some females, energy intake does not match estimated energy expenditure from exercise (Beidleman et al, 1995; Cumming et al, 1994; Loucks, 1996). A chronic state of such may result in the body becoming more efficient to preserve energy necessary for life function, including thermoregulation, cellular maintenance and locomotion (Wade et al, 1996). Therefore, in these athletes suppressed physiological adaptations may present (Loucks et al, 1994b; Wade et al, 1996).

Studies that have measured components of energy balance in relation to reproductive physiology in females and males will be reviewed. Bullen et al (1985) employed an 8-week program of strenuous exercise in previously untrained females. Weight-loss occurred in one group while weight was maintained in the other by adjusting energy intake. Although both groups had disruptions to the menstrual cycle, the prevalence was lower in the weight-maintenance group. Habitual energy intake is often similar between amenorrheic and eumenorrheic endurance-trained athletes as well as non-athletic controls despite significantly higher duration of training per week in the amenorrheic group (Harber et al, 1998). Amenorrheic athletes also displayed significantly lower T_3 levels compared to eumenorrheic athletes and controls in this study (Harber et al, 1998) and suggests an association between insufficient energy intake, increased energy expenditure and alterations to reproductive hormone profiles. In males, insufficient energy intake during an extensive 95-day expedition coincided with a marked decrease in testosterone and mean LH values. Negative energy balance was suggested to have developed in these

individuals (Stroud et al, 1997). Conversely, eight elite athletes participated in a 7-day study wherein they consented to have energy intake recorded and their energy turnovers measured through use of doubly-labeled water (Sjodin et al, 1994). Positive energy balance (defined by authors as energy intake minus energy turnover) occurred over the 7 days however there were no endocrine measures taken. The extent to which the presence of a chronic state of negative energy balance exists in athletes is unclear.

3.2 Eating attitudes

Anorexic tendencies are associated with suppressed hormones of the reproductive system as well as menstrual cycle irregularities (Fichter et al, 1989). Although anorexia nervosa is less prevalent in males, Wheeler et al (1986) noted an increased tendency towards aberrant eating attitudes accompanied by lower levels of testosterone in high mileage runners in comparison to low mileage runners and sedentary males. Allotting time to train over to eat was a regular occurrence for 18% of male runners and an occasional occurrence for 55% (Wheeler et al, 1986). Restrained eating is associated with reduced daily energy intake in males (Tepper et al, 1996) however, the extent to which eating attitudes in high endurance athletes may affect energy intake or reproduction in males is unknown. Examination of eating attitudes in male athletes is extremely limited, therefore conclusions cannot be drawn regarding differences of eating attitudes existing between male athletes and non-athletes.

3.3 Energy intake

There is much information regarding energy intake and the H-P-G axis in females but there are limited data describing energy intakes of the male athletic population. Both moderate and severe energy restriction have been studied with respect to female reproductive hormones and suggest that, in females, acute depression of energy intake results in disruptions to the H-P-G axis (Cameron, 1989; Loucks, 1996; Loucks et al, 1994a; Olson et al, 1995; Rosetta, 1993; Williams et al, 1995). Sedentary females experienced a decrease in LH pulse frequency with an increase in LH pulse amplitude with 4 days of restricted energy intake (10 kcal/kgLBM/day) (Loucks et al, 1994b). A three-day fast in females resulted in significantly decreased LH pulse frequency with no accompanying change in LH mean value or amplitude nor subsequent follicular development or ovulation (Olson et al, 1995). Anorexia nervosa patients, who are in a chronic state of caloric restriction, often have LH patterns resembling of those of young children (Fichter et al, 1989). These patterns of LH are of insufficient amplitude to elicit an LH surge, thus impacting the menstrual cycle.

Research on the effects of energy deficiency on male reproduction is limited, however acute responses of the hormonal axis to energy deprivation may provide insight to the mechanisms responsible. Cameron et al (1991) reported decreased testosterone, LH and FSH concentrations in healthy men within 48 hours of fasting. LH pulse frequency and mean value concentrations were suppressed, but amplitude was unaltered. Cortisol was also measured but unchanged, which suggested that fasting was not associated with an adequate stress to the adrenal axis and therefore the

dampening of the reproductive axis may be independent of the adrenal axis under these circumstances. Short-term fasting inhibited Leydig cell function as testosterone levels decreased by 34 % (Rojdmark, 1987). As blood glucose levels dropped significantly during the study, a second group was administered oral glucose supplementation during the same fasting period. The ingestion of the energy from the oral glucose supplement resulted in no decrease in testosterone as previously experienced with the fast (Rojdmark, 1987). The decreased secretion of testosterone in response to decreased energy intake may be attributed to LH responses as has been demonstrated in females in fasting environments. Weight fluctuation, imposed by alterations in energy intake, may also affect male reproductive hormones. Wrestlers, who need to reduce body weight during their competitive season, may be at risk of suppressed levels of circulating reproductive hormones. Male wrestlers often have decreased body fat and body weight during their in-season accompanied by decreased testosterone and prolactin in comparison with their post-season states (Strauss et al, 1985). There was a strong positive correlation between percent body fat and serum testosterone in the amateur wrestlers during their competitive season.

There has been minimal research on the effects of dietary components on physiological changes to the reproductive system in males. De Souza et al (1994a) analyzed 4-day habitual diet records and found no significant differences between males other than increased carbohydrate consumption in the high mileage runners compared to the moderate mileage runners and sedentary controls. Macronutrient intake of male endurance athletes whose energy consumption was considered "low" (negative energy balance) was compared to those whose intake was considered

"adequate" (positive energy balance). There was no difference in percent of fat, protein or carbohydrate intake between the groups (Thompson et al, 1995). However, when expressed as grams/day or grams/kilogram body weight, fat and protein were lower in the low energy group (negative energy balance) (Thompson et al, 1995). Further research is necessary to examine the effects of energy and macronutrient intake on reproductive hormone profiles in males.

3.4 Resting energy expenditure

Resting energy expenditure (REE) is a major contributor to total energy expenditure and is important when analyzing energy balance. Prediction formulas are often used as a means of estimating energy expenditure at rest. Insight to the influence of energy restriction as well as energy expenditure on REE may contribute to greater understanding of the role of REE in energy balance.

Inadequate energy intake for males who have high training volumes may result in a state of negative energy balance. The outcomes of this imbalance on REE are inconclusive. It had been hypothesized that resistance training would preserve REE (absolute) through increase in fat free mass during a weight-loss program in obese subjects (Geliebter et al. 1997). Compared to the other groups, who exercised aerobically or relied on diet alone for weight loss, there were no significant differences with respect to REE after the 8-week program. Ballor et al (1995) performed a meta-analysis on REE reported in previous studies. Results indicated that weight-loss from diet restriction had negative effects on REE while exercise during weight-loss had no significant effects.

Observed increases in REE (absolute) in athletes may be attributed to acute effects of exercise on metabolism rather than a chronic adaptation of the metabolic system (Bullough et al, 1995). It has been suggested that athletes with higher REE may be less efficient during non-exercise portions of the day compared to sedentary controls (Beidleman et al, 1995; Burke et al, 1993). However, much research suggests that the REE of endurance athletes may not be different or, conversely, slightly lower in comparison to non-athletes (Thompson et al, 1995; Tremblay et al, 1997). Where highly trained male endurance athletes of matched energy expenditure recorded caloric intake for 7 days, 6 of 10 participants were classified as having low energy intake, resulting in negative energy balance (Thompson et al, 1995). This group displayed significantly lower 24-hour energy expenditure (absolute), primarily accounted for by resting energy expenditure and sleeping energy expenditure. Endurance training resulted in an 8% decrease in REE (absolute) in males, even though fat-free mass was preserved (Tremblay et al, 1997). The effect of energy balance on REE may support the concept of conservation of energy when availability is compromised (Drinkwater et al, 1984; Myerson et al, 1991). This conservation could result in decreased REE accompanied by suppressed reproductive hormones as a means of maintaining energy for basic life function (Cumming et al, 1994; Loucks et al, 1994b; Wade et al, 1996).

3.5 Thermic effect of feeding

Thermic effect of feeding refers to the energetic demands on the body for ingesting and digesting foods. The average thermic effect of feeding (TEF) for adults is approximately 10 % of total daily energy expenditure (McArdle et al, 1991). Witt et al (1993) identified trained males to have significantly higher TEF than untrained males, independent of carbohydrate ingestion. Other research has measured TEF through indirect calorimetry following the ingestion of a known macronutrient liquid meal and found no significant differences between trained and untrained females (Beidleman et al, 1995; Burke et al, 1993). Although there is no literature addressing TEF with respect to hormonal profiles of the reproductive axis in males, it should be noted that it is a component of energy expenditure and should therefore be included in estimation of total daily energy expenditure.

3.6 Exercise energy expenditure

3.6.1 Volume of training and alterations of reproductive physiology

Extensive information is available regarding volume and intensity of activity in both male and female athletes as they pertain to physiological adaptations of the reproductive system, including suppressed hormones. In females, an exercise threshold at which menstrual cycle irregularities occurs in females has been defined using volume and intensity of exercise (Beitins et al, 1991; Bullen, 1984; Bullen et al, 1985; Rogol et al, 1992). More recently, a similar exploration of volume of exercise in male athletes has been completed. Volume threshold was defined as increased volume of endurance training and was associated with suppression of male

reproductive hormones as well as subclinical changes to semen quality (De Souza et al, 1994a). Although a specific volume of training was not identified where the male reproductive hormonal profile and, or semen characteristics were altered physiologically, the following categories were identified. High mileage runners defined as running a minimum of 104 kilometers per week (9.7 hours per week) had significantly lower total testosterone and free testosterone than moderate mileage runners (40 to 56 kilometers per week, averaging 3.2 hours per week) as well as sedentary controls (De Souza et al, 1994a). High mileage runners displayed lower motile sperm count, sperm density and penetration capabilities than controls. Sperm morphology was also significantly altered in the high mileage group compared to the moderate group or the controls. De Souza et al (1994a) found a significant correlation between these semen characteristics and volume of training (km) where r = 0.375 for semen volume, r = 0.484 for sperm motility, r = 0.376 for sperm density and r = 0.485 for sperm morphology (round cells). Habitual energy intake differed, when expressed as kcal/kg/day, in that high mileage runners consumed more than moderate or controls. However, it was not calculated as to whether the intake was sufficient compensation for expenditure of energy from the high mileage athletes. Conclusions were drawn that a threshold of training existed, above which male gonadal hormone and sperm production was suppressed. A similar threshold has been proposed in male athletes presenting with low testosterone with no change to LH, suggesting possible interruptions to the reproductive axis from exercise may be limited to the gonads (McColl et al, 1989). Investigation of other characteristics in a high mileage population, including exercise energy expenditure, energy intake, eating

attitudes and resting energy expenditure were not measured (McColl et al, 1989). Further discussion on exercise energy expenditure through exercise will follow in the next section.

Volume and, or mileage of training, especially in runners, has been consistently correlated with lower total testosterone (Arce et al, 1993b; Hackney et al, 1990; Hackney et al, 1988; McColl et al, 1989; Wheeler et al, 1991; Wheeler et al, 1986; Wheeler et al, 1984). Free testosterone, the biologically active form of the gonadal sex steroid, was significantly decreased in endurance trained males (Arce et al, 1993b; Hackney et al, 1988; Wheeler et al, 1991; Wheeler et al, 1986; Wheeler et al, 1984). Further, there have been reports of decreased LH pulse frequency, decreased area under the LH curve (MacConnie et al, 1986; McColl et al, 1989) and changes in sperm quality (Arce et al, 1993b; Jensen et al, 1995). Exhaustive endurance training and over-trained states result in similar findings (Griffith et al, 1990; Roberts et al, 1993).

3.6.2 Exercise expenditure and the H-P-G

Two weeks of exhaustive endurance training (cycling and running) resulted in decreased plasma testosterone accompanied by a decline in sex drive and sexual activity (Griffith et al, 1990). The lower sex drive may have been a symptom of reproductive dysfunction, may have been from general fatigue or may have been a sign of overtraining (Griffith et al, 1990). However, as sex drive is a CNS response, it is possible that neuroendocrine stimulus at the hypothalamus may be affected with endurance exercise.

In females, routine training has been associated with a decrease in LH pulse frequency and increase in pulse amplitude (Cumming, 1989a; Loucks et al, 1989). The findings in males are less conclusive. Six male marathoners presented with decreased frequency and amplitude of LH compared to healthy controls (MacConnie et al, 1986). Gonadal hormones did not differ at baseline, but increased in response to an exercise bout. Temporary elevated concentrations of these hormones resulting from daily exercise may lead to suppression of GnRH. A deficiency in hypothalamic GnRH may have been present in male marathon runners (MacConnie et al, 1986). Blunted LH response to exogenous GnRH administration in endurance-trained males may have been indicative of alterations to the H-P-G axis not present in sedentary males (Hackney et al, 1990). Hormonal profiles of male athletes, with a training volume greater than 80 kilometers per week, revealed depressed LH levels without an effect on pulsatile release (McColl et al, 1989). In contrast to these findings, some find no differences in LH (Arce et al, 1993b; Wheeler et al, 1991) or an increase in LH (Hackney et al, 1990). Prolactin, as well, has been noted as reduced (Wheeler et al, 1984; Wheeler et al, 1991), unaltered (Arce et al, 1993b; Hackney et al, 1988) as well as elevated (Jensen et al, 1995) with endurance training.

Although methodological inconsistencies with respect to sampling for LH pulsatility can not be overlooked, evidence suggests that volume of endurance training may have an effect on hormonal changes at the levels of the gonads and the pituitary. The mechanisms for observed hormonal differences are not fully understood. It has been suggested that a threshold of exercise may exist where testosterone is altered at a certain volume of training, and a much greater volume is

necessary to evoke changes in LH (McColl et al, 1989). This would support data reporting decreased chronic testosterone response to endurance exercise with no alterations to the hypothalamic or pituitary hormones (Hackney et al, 1990; Wheeler et al, 1991; Wheeler et al, 1984).

In a cross-sectional study, both total and free testosterone were lower in runners than untrained males while prolactin and cortisol were not different (Hackney et al, 1988). LH was higher in these runners, reducing the possibility that the H-P-G axis was directly responsible for alterations to gonadal secretion of anabolic steroid hormones. The authors suggest that testicular function may have been altered and therefore related to the observed decrease in testosterone production. Further to this, a short-term over-training protocol in 5 endurance-trained males resulted in a significant decrease in testosterone and sperm concentration (Roberts et al, 1993). Three months post-training, testosterone levels had returned to baseline however sperm concentration remained significantly lower. This suggests that testicular function was altered, as previously proposed (Hackney et al, 1988). Although the previous studies were performed in runners, there are indications that exercising on a bicycle may directly affect the testes. It has been suggested that temperature control and, or microtrauma to the testes while cycling may partially accounted for noted alterations to sperm motility in competitive cyclists that were not observed in marathon runners (Lucia et al, 1996).

3.6.3 Methodological considerations of measuring energy expenditure

Volume of training, used as a measure for energy expenditure, may be essential information in evaluating the reproductive system in athletes. Failure to differentiate high endurance athletes from recreational athletes may result in no significant differences with respect to endocrine profiles. Comparison of endurance-trained males and resistance-trained males with untrained males resulted in alterations to reproductive hormonal profiles of athletes yet only endurance-trained males displayed altered sperm characteristics (Arce et al, 1993b). Therefore, details regarding training parameters (volume, duration, intensity, frequency) are needed when analyzing physiological changes to the reproductive system in male athletes. As such changes are often subclinical in males, appropriate attention regarding training volume may provide identification of males at risk of fertility complications. Therefore, it is imperative that training volume is better understood with respect to changes in reproductive physiology.

Few studies have accurately calculated energy expenditure from exercise. Speed of training may provide indication of training intensity, as would heart rate records however few studies have reported this information. Monitoring physical activity through self-reported measures are common methods for quantifying energy expenditure from exercise. Training volume, expressed as distance or hours per given time period, are generally reported. Ideal methods of closed chamber calorimetry are not practical in many sport settings, however accelerometers such as the TriTrac R3-D™ and CalTrac™ monitors are both valid and reliable for extrapolating information regarding energy costs of movement (Welk et al, 1995).

There has been limited research on the effects of intensity of training or training modalities other than running with respect to energy expenditure and the male reproductive system. Lucia et al (1996) compared hormones and semen characteristics of competitive cyclists with triathletes, runners and controls across different periods of the training calendar. There were no significant differences in reproductive hormones or sperm quality, with exception of the cyclists portraying decreased sperm motility during the competitive season. Training characteristics for the different athletes were not well-described and energy expenditure was not measured. Neary et al (1994) also investigated hormonal alterations to training in male cyclists. However, most research has examined runners, therefore transferring these findings to other sports may be problematic.

4. SUMMARY

There is sufficient research to suggest that energy costs of endurance exercise may interrupt reproductive function in both females and males. Although insufficient energy intake for total energy expenditure may increase risk of suppressed reproductive hormone profiles, calculations of energy balance as they relate to such are limited. To date, a volume threshold effect has been identified in males, where exceeding the threshold with respect to volume of training, is associated with a decrease in circulating testosterone as well as alterations to spermatogenesis (De Souza et al, 1994a). Reduced energy availability has also been explored in females with respect to menstrual cycle properties (Loucks et al, 1994b). There is sufficient

evidence to suggest that the male H-P-G axis is affected in a similar manner to females, hence this information may relate to the male athletic population. Information on energy intake has indicated that acute bouts of fasting may suppress pituitary and gonadal endocrine production (Cameron et al, 1991; Rojdmark, 1987). Chronic effects of inadequate energy intake on the male reproductive system have not been measured. Eating attitudes and their influences on eating behaviors in male endurance athletes has not been measured in evaluating energy balance or suppressed reproductive hormones. Where much attention has been focused on volume of training, direct measurement of energy expenditure from exercise has not occurred. Further, most research on exercise and chronic hormonal changes have been limited to running as the primary modality.

Characteristics of highly trained endurance male athletes, who have disruptions to the H-P-G axis in comparison to athletes of moderate endurance and sedentary males, have not been clearly identified. Energy balance, energy intake, eating attitudes, resting energy expenditures and exercise energy expenditure need further investigation in endurance male athletes and sedentary individuals to determine the effects of these variables on the H-P-G axis.

CHAPTER 3

METHODS

1. EXPERIMENTAL DESIGN

The experimental design was a descriptive cross-sectional study consisting of three groups of young men. All groups completed similar measurements and questionnaires. Ethical approval was obtained from the Faculty of Physical Education and Recreation at the University of Alberta (Appendix A).

2. SUBJECT INFORMATION

Fifty-two healthy males, including athletic and sedentary individuals, volunteered to participate in this investigation. Athletes were engaged predominantly in cycle training. Subjects were recruited through word of mouth, posters at the University of Alberta campus and various cycling stores in Alberta as well as an advertisement in a provincial cycling newsletter. Samples of the recruitment posters are included in Appendix B. Upon completion of an informed consent (Appendix C). subjects were assigned to one of three groups depending upon their self-selected number of hours of aerobic training per week (Appendix D, Section A, Question 1) (Table 2). Each individual was interviewed at the time to confirm selection based upon yearly training schedule. Sample size for each group was 16 in the High Endurance (HE), 20 in the Moderate Endurance (ME) and 16 in the Sedentary Control (SC).

Table 2. Group categories determined by self-selected number of hours of training

Group	Self-selected Hours of Aerobic Training / Week
High- Endurance (HE)	**11 – 15 +**
Moderate Endurance(ME)	**5 – 10**
Sedentary Control (SC)	**0 – 4**

The following eligibility criteria were met by each subject:

1) Chronological age of 20 to 38 years.
2) Healthy individuals free from injury, with no reported history of thyroid or eating disorders, and free from use of medications within the last 6 months that are known to influence hormonal status.
3) Non-smoking.
4) Stable body weight over the last 6 months (± 3 kg); body fat < 26%, determined by hydrostatic weighing.
5) Intense regular resistance training was not a component of activity program. Resistance training was not performed more than 2 times per week and weight lifted was less than 75% of maximal effort.

3. SCREENING

3.1 Health Status and Reproductive Profile: Each subject completed a demographic questionnaire (Appendix D) that provided information about health status, medical history, use of medications, known reproductive problems or complications with regards to reproductive or metabolic hormones.

3.2 Physical Activity Status: The completed demographic questionnaire (Appendix D) provided information about subjects' training volume including modality, intensity, duration and frequency for the past 12 months as well as level of competition, if relevant. Subjects were asked to record the number of hours of aerobic exercise completed each week over 12 months. Each subject also documented his current training routine (where applicable). This information was used as a confirmation of hours of aerobic training per week as well as provides descriptive information regarding modality, intensity, duration and frequency of exercise in each group.

4. MEASUREMENT PROTOCOLS

A schematic diagram of the study protocol is illustrated in Figure 3.

4.1 Resting Energy Expenditure: Subjects reported to the Metabolic Testing Lab in the Faculty of Agricultural, Food and Nutritional Sciences at the University of Alberta between 7:00am and 9:00am. No exercise was permitted 24 hours prior to testing and subjects were fasted for 12 hours. Subjects were asked to arrive at the lab at a leisurely pace where they arrived by motor vehicle and took the elevator to the 4^{th} floor to the Metabolic lab. Following an explanation of the test, each subject rested for 30 minutes in a supine position while lights were dimmed and relaxation music was played. A transparent ventilated hood was then placed over the subject while oxygen consumption was measured at rest for another 30 minutes. Subjects were not permitted to sleep or move during the 30 minutes of steady state measurements. Oxygen consumption was measured by indirect calorimetry using a

metabolic cart (Vmax 29N, Sensormedics, Yorba Linda, California). The metabolic cart was calibrated against a reference gas mixture and relevant information including height, weight, age, gender were entered into the metabolic software. Steady state values were for minute ventilation, VO_2 and respiratory exchange ratio (RER). The Weir equation was programmed into the computer to calculate resting energy expenditure (REE) (kcals/day). The Weir equation is as follows:

$$\textbf{REE (kcals/day)} = 3.9[VO_2(ml/min)] + 1.1[VCO_2(ml/min)] \times 1.41$$

4.2 Body Composition: This was established through two methods of assessment: sum of five skinfolds (CSTF, 1986) and hydrostatic weighing (Brozek et al, 1963) using a helium dilution method for determining residual volume (Motley et al, 1957). Both sum of skinfolds and hydrostatic weighing were measured on the same day. All measurements were taken at the University of Alberta, Faculty of Physical Education and Recreation by the same technician. Subjects were asked to refrain from eating 4 hours prior to testing as well as to refrain from exercise, prior to testing. Skinfold sites included biceps, triceps, subscapularis, iliac crest and medial calf. Measures were taken twice and measurements of 0.4mm or less were averaged. A third measure was taken where values exceeded 0.4mm. The two closest measures were then averaged to determine individual site measures.

Residual volume was measured with the helium dilution technique on a SensorMedics 2450, Pulmonary Function Laboratory cart (Yorba Linda, California). Test procedures were explained to each subject.

4.3 Aerobic Fitness Assessment: Maximal aerobic consumption (VO_2max) was determined using an incremental protocol on a Monarch cycle ergometer (Varberg, Sweden). Subjects maintained a cadence not less than 60 rpm with increased resistance of 0.5 kp every 2 minutes until ventilatory threshold was indicated by a decrease and plateau in Ve/VCO_2 prior to a systematic increase with increased power output as well as a respiratory exchange ratio greater than 1.05 (Bhambhani et al, 1985). Resistance was then increased every minute by 0.5 kp to volitional exhaustion. Gases were analyzed every 20 seconds using an open circuit spirometer (SensorMedics Horizon Metabolic Cart, Anaheim, California). Heart rate was recorded every minute (Polar Electro Heart Rate Monitor, Polar USA Inc, Stanford, Connecticut). Criteria for peak VO_2 included: 1) no increase in O_2 uptake greater than 100 ml with increased exercise intensity (plateau criteria); 2) a respiratory exchange ratio greater than 1.1; 3) volitional exhaustion (Thoden, 1991).

4.4 Dietary Intake: Subjects were trained by the investigator to complete a 4-day dietary intake record. All details of food consumption were recorded in the booklets provided. Food labels and nutritional information sheets were also collected when possible. A sample of the dietary record sheet and hint sheet is provided in Appendix E. The 4-day diet collection coincided with the 4-day physical activity assessment to assist with energy balance determination. Diet records were analyzed using the Food Processor II ™ program (ESHA Research, Oregon). Accuracy checks were performed after the 4-day recording to ensure food measurements and food descriptors were detailed and accurate. The analyzed dietary intake record provided

information regarding habitual energy intake and macronutrient sources. Averages were obtained for total energy, carbohydrate, fat and protein intakes (g and % of total daily energy intake).

4.5 Physical Activity Assessment:

4.5.1 TriTrac R3-D™: Subjects wore a portable accelerometer, the TriTrac R3-D™ calorimeter monitor (Hemokinetics, Madison, WI, 1996) on their hip in a special carrying case for 4 days other than while sleeping or engaged in water activities (ie. showers or swimming). Subjects' weight, height, age and gender were entered into the computer software program and the TriTrac R3-D™ was programmed to record motion at 1-minute intervals. Once the TriTrac R3-D™ was loaded with the subjects' information, the subject was instructed to wear the unit for the specified 4 days. The TriTrac R3-D™ measured activity on three planes: horizontal (y-axis), vertical (x-axis) and mediolateral (z-axis). It calculated and displayed resting energy expenditure, energy expenditure from exercise as well as total energy expenditure. Data were recorded in the TriTrac R3-D™ unit for these days and was then downloaded into the software program to display energy expenditure. The equation used to determine energy expenditure from exercise was based on the body mass multiplied by acceleration principle where vector magnitude was determined for each minute from the three planes measured. The equation is as follows:

$$\textbf{Vector Magnitude} = \sqrt{(\text{x-axis})^2 + (\text{y-axis})^2 + (\text{z-axis})^2}$$

(Hemokinetics, Madison, WI, 1996)

The equation for calculating metabolic calories (resting energy expenditure per minute) for men is as follows:

$$\textbf{Kcals/min} = \frac{[473 \times \text{weight (kg)}] + [971 \times \text{height (cm)}] - [513 \times \text{age (years)}] + 4687}{100\,000}$$

(Hemokinetics, Madison, WI, 1996)

The TriTrac R3-D™ provides a reliable and valid measurement of energy expenditure in daily activities (Welk et al, 1995). The TriTrac R3-D™ has not been validated for cycling activity.

4.5.2 Physical Activity Logbook: For the same 4 days, subjects recorded all bouts of physical activity in a detailed logbook (Harber et al, 1998). A copy of this logbook is provided in Appendix F. Subjects were asked to continue their usual level of physical activity for the four days. They were required to record their activities, the number of minutes they participated in the activities as well as the perceived intensities of the activities. Perceived intensities from which to choose included: "1" (Very light, not vigorous at all), "2" (Light, somewhat vigorous), "3" (Medium, moderately vigorous), "4" (Heavy, vigorous) and "5" (Very very heavy, extremely vigorous). Duration of exercise at various intensities were recorded. The endurance athletes were provided an additional recording sheet for all cycling activity. This form was more detailed in that it required subjects to record distance traveled and average speed while cycling which was determined through using a cyclocomputer (provided by researcher where necessary) as well as intensity and duration of exercise.

4.6 Eating Attitudes: Subjects completed the Eating Disorder Inventory (EDI) (Garner et al, 1983) and the Dutch Eating Behaviour Questionnaire - Restraint Subscale (DEBQ-R) (van Strien et al, 1986). The EDI questionnaire consisted of 64 questions including 8 subscales (drive for thinness, bulimia, body dissatisfaction, ineffectiveness, perfectionism, interpersonal distrust, interoceptive awareness and maturity fears). Test-retest reliability of the EDI subscales has been demonstrated in both males and females (Garner, 1991). The DEBQ-R has been reported as both reliable and valid in males and females (van Strein et al, 1986; Allison, 1995). These questionnaires were coded to establish anonymity. Questionnaires were completed following the 4-day diet and activity records. A sample of questions from these questionnaires is provided in Appendix G.

4.7 Serum Hormonal Concentration

4.7.1 Blood collection: A 20 ml blood sample was obtained from each subject at the anticubital region by trained personnel. Subjects were requested to arrive between 4 pm and 6 pm and rest for 15 minutes prior to sample being drawn to account for postural plasma shifts. They were requested to refrain from exercise that day prior to the blood sample being drawn. Subjects were asked to keep well hydrated throughout the day and requested to refrain from eating for 4 hours prior to sample collection. Samples were allowed to clot at room temperature for 45-60 min and then centrifuged at 4°C for 12 minutes at 2500 revolutions per minute. Serum was aliquotted and then stored at -80°C until RIA analysis

4.7.2 Analysis of blood: Serum was analyzed for free testosterone, total testosterone, cortisol, FSH, LH, Prolactin, T3 and T4. Serum samples were allowed to thaw one time and were then processed using radioimmunoassays (RIA) using reagent kits (Coat-A-Count, Diagnostic Products Corporation, Los Angeles, CA). All samples were analyzed in duplicate within a single assay according to the instructions provided by the manufacturers of the kits. Controls with known concentrations were added to each assay to determine intra-assay variation. Precision of each assay was measured by a computer-generated coefficient of variability (CV). Acceptable intra-assay CV's were 6 - 15% (Chard et al, 1987).

5. CALCULATION OF ESTIMATED TOTAL ENERGY BALANCE

Energy balance was derived from calculating total energy expenditure and subtracting that from total energy intake. Total energy intake was obtained from the average of the 4-day dietary intake (section 4.4). Total energy expenditure was calculated by combining resting energy expenditure, energy expenditure from exercise and thermic effect of feeding. Resting energy expenditure (REE) was estimated from the results of REE test at the Metabolic Testing Lab at the University of Alberta (section 4.1). Energy expenditure from daily active living and exercise was estimated from the average of the 4-day activity portion of results determined by the TriTrac R3-D (section 4.5.1). Thermic effect of feeding was estimated from calculating 10% of REE (McArdle et al, 1991, Melby et al, 1998). The product of the equation reflects estimated total energy balance where a negative value would reflect

negative energy balance and would suggest that energy intake may be insufficient for total energy expenditure.

6. STATISTICAL ANALYSIS

One way ANOVA's were performed on all dependent variables for each of the three groups. Statistica 5.1 software program (Statsoft Inc., 1997, Tulso, OK) was used for all statistical analysis. An alpha of less than 0.05 was used to establish significance. Post-hoc analysis, using Neuman-Keuls, was conducted to determine differences between groups, when necessary.

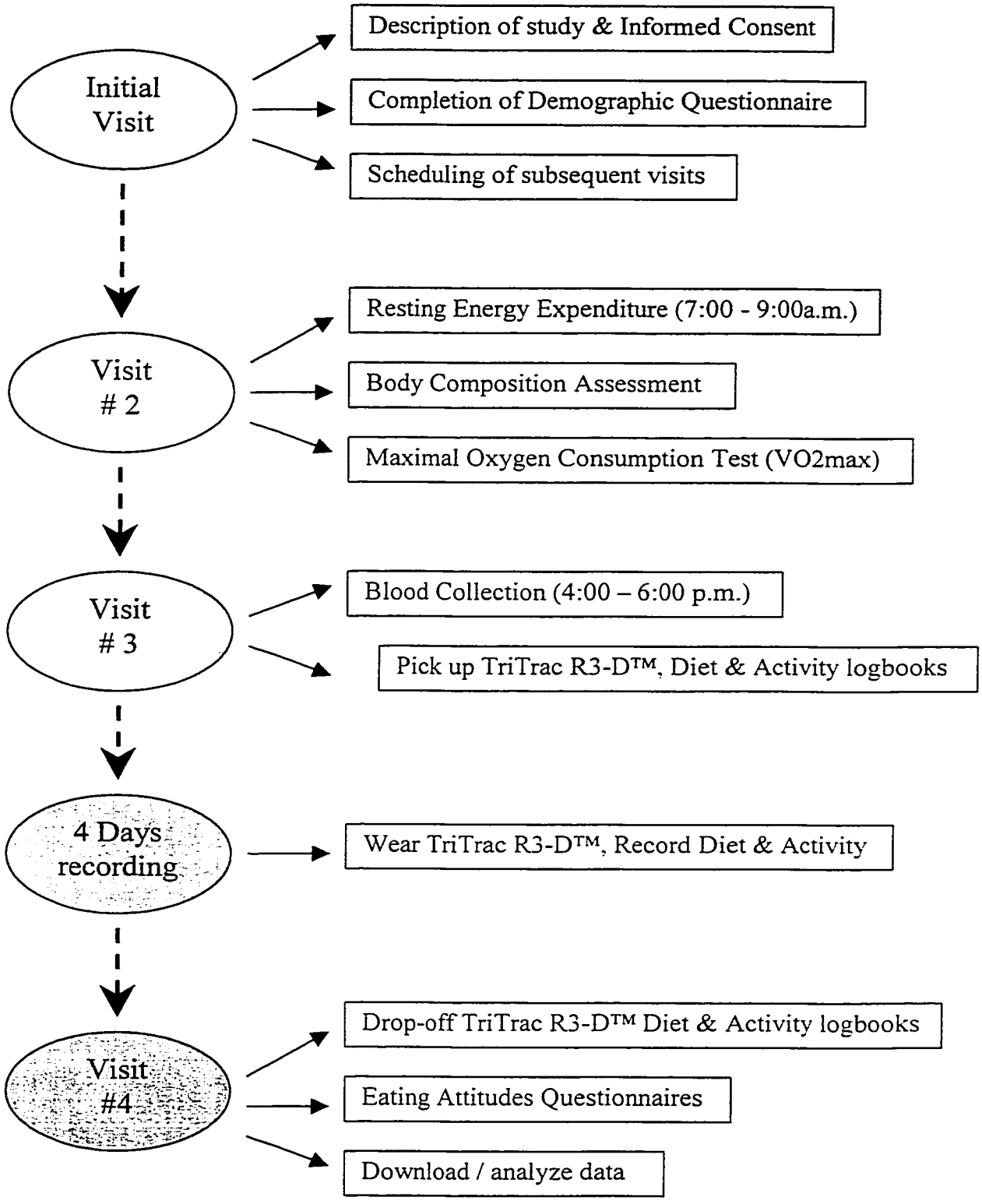


Figure 3. Schematic diagram of study protocol.

CHAPTER 4

RESULTS

1. Demographics and groupings

Fifty-two males completed the requirements of this study. All subjects reported being in good health and reported having no known reproductive problems or complications with regards to reproductive or metabolic hormones. Two subjects reported using medications or nutritional supplements but terminated their use at least 4 weeks prior to commencing the study.

Athletes selected for this study were engaged predominantly in cycle training. This included competitive road cyclists, mountain bikers and triathletes. Subjects were grouped through a self-selected aerobic activity question on a demographic questionnaire. Subjects identified their regular hours of aerobic training per week on a yearly basis on a multiple choice question (Appendix D, Section A, Question 1). This answer was used to distinguish groups. The high-endurance group (HE) was defined as greater than 10 hours of aerobic training per week on the multiple choice question. The moderate-endurance group (ME) was defined as 5 to 10 hours of aerobic training per week. The sedentary control group (SC) was defined as less than 5 hours per week of aerobic exercise. Further details pertaining to specific current training regiments were recorded in detail on the questionnaire (Appendix D, Section A, Question 7b) and results are shown in Table 3. There were significant differences with respect to hours of current weekly aerobic training.

Table 3 - Duration of self-reported detailed weekly aerobic activity. [**mean**, ± SD, (range)]

Group	Sample size	Reported aerobic activities* (hours/week)
HE	16	**16.0** ±5.4 (8 – 28.5)
ME	20	**8.6** ±1.9 (5.5 – 12)
SC	16	**1.4** ±1.3 (0 – 4)
p value	-	.000117[a] .000117[b] .000126[c]

Note: HE = High-endurance male cyclists
ME = Moderate-endurance male cyclists
SC = Male sedentary controls

a = ME significantly different than SC
b = HE significantly different than ME
c = HE significantly different than SC

*Reported aerobic activities = these data were taken from demographic questionnaire pertaining to detailed current specific training question (Appendix D, Section A, 7b).

2. **Subject characteristics** (Table 4)

There were no differences in age (years) between the three groups. Height (cm) was significantly lower in ME compared to HE, while weight (kg) was significantly lower in ME than SC. Body composition assessment resulted in significantly lower sum of skinfolds (mm), percent body fat as well as fat mass (kg) in both HE and ME in comparison to SC. Fat free mass (kg) was not different between groups.

3. **Maximal aerobic consumption and ventilatory threshold** (Table 5)

Maximal aerobic consumption, expressed in litres per minute, was significantly different between all three groups. HE was significantly higher than ME and SC and ME was also significantly higher than SC. Maximal aerobic

consumption, expressed as either ml/kg/min or ml/kgFFM/min, resulted in HE and ME significantly higher than SC with no significance between HE and ME. Ventilatory threshold was identified for each group and HE was significantly higher than ME and SC and ME was significantly higher than SC.

Table 4 - Subject characteristics. [**mean**, ± SD, (range)]

Group	Age (years)	Height (cm)	Weight (kg)	Sum of Skinfolds (mm)	Percent Body Fat (%)	Fat Mass (kg)	Fat Free Mass (kg)
HE (n = 16)	**27.7** ±5.1 (22 – 38)	**181.8** ±5.0 (173 – 193.2)	**77.3** ±7.8 (66.6 – 90.4)	**38.3** ±9.5 (26.4 – 65.05)	**12.4** ±2.3 (7.8 – 17.4)	**9.6** ±1.9 (6.8 – 13.1)	**67.8** ±7.3 (58.2 – 81.2)
ME (n = 20)	**27.0** ±5.2 (20 – 38)	**176.2** ±6.0 (164.2 – 187.8)	**71.9** ±10.3 (55.5 – 98)	**42.8** ±18.8 (24.9 – 71.65)	**12.8** ±5.0 (6.1 – 26.7)	**9.5** ±5.2 (4.5 – 26.2)	**62.6** ±6.5 (49.8 – 74.9)
SC (n = 16)	**26.9** ±4.4 (21 – 35)	**180.0** ±7.5 (173.6 – 192.3)	**82.4** ±9.8 (62.4 – 97.4)	**66.2** ±10.7 (48.4 – 84.65)	**19.9** ±4.6 (14.5 – 27.7)	**16.4** ±4.4 (10 – 25)	**66.1** ±8.5 (53.4 – 80.9)
p values	.890556	.030836[b]	.005909[a]	.000126[c] .000128[a]	.000135[c] .000125[a]	.000130[c] .000162[a]	.107871

Note: a = ME significantly different than SC
b = HE significantly different than ME
c = HE significantly different than SC

Table 5 - Maximal oxygen consumption (VO_2max) and Ventilatory threshold. [**mean**, ± SD, (range)]

Group	VO_2max (l/min)	VO_2max (ml/kg/min)	VO_2max (ml/kgFFM/min)	Ventilatory Threshold (l/min)
HE (n = 16)	**4.8** ±0.5 (3.6 – 5.5)	**62.0** ±5.7 (52.3 – 72.2)	**71.0** ±6.29 (60.1 – 80.7)	**3.9** ±0.6 (3.1 – 4.8)
ME (n = 20)	**4.2** ±0.6 (3.3 – 5.3)	**59.3** ±6.5 (43.7 – 70.7)	**68.0** ±6.4 (57.6 – 77.7)	**3.4** ±0.5 (2.5 – 4.3)
SC (n = 16)	**3.5** ±0.6 (2.5 – 4.6)	**42.6** ±5.9 (31.8 – 51.4)	**53.2** ±5.8 (44.1 – 63.5)	**2.7** ±0.5 (1.8 – 3.5)
p values	.004901[b] .000126[c] .000343[a]	.000126[c] .000117[a]	.000126[c] .000117[a]	.005276[b] .000126[c] .000268[a]

Note: a = ME significantly different than SC
b = HE significantly different than ME
c = HE significantly different than SC

4. 4-day energy intake (Table 6)

HE and ME had significantly higher 4-day average energy intake than SC when expressed as kcal/day and kcal/kgFFM/day. Percent fat intake was significantly higher in SC than ME whereas carbohydrate and protein did not differ between groups. Appendix H contains the daily diet record information for each group, including kcal/day, kcal/kg FFM/day, calories from fat (% and g), calories from carbohydrate (% and g) and calories from protein (% and g).

Table 6 - 4-day dietary intake. [**mean**, ± SD, (range)]

Group	Energy intake (kcal/day)	Energy intake (kcal/kgFFM/ day)	Calories from Fat (%)	Calories from Carbohydrate (%)	Calories from Protein (%)
HE (n = 16)	**3470** ±523 (2718 – 4471)	**52** ±8 (42 – 62)	**25** ±7 (12 – 35)	**59** ±9 (48 – 77)	**15** ±2 (11 – 20)
ME (n = 20)	**3445** ±499 (2523 – 4464)	**55** ±8 (40 – 72)	**25** ±6 (12 – 36)	**58** ±8 (44 - 71)	**14** ±2 (11 – 17)
SC (n = 16)	**2709** ±513 (1771 – 3390)	**42** ±9 (27 – 56)	**30** ±5 (22 – 43)	**53** ±7 (40 – 62)	**15** ±3 (11 – 20)
p values	.000306[c] .000216[a]	.001418[c] .000173[a]	.046459[a]	.082276	.652784

Note: a = ME significantly different than SC
c = HE significantly different than SC

5. Resting energy expenditure (Table 7)

Oxygen consumption (l/min) at rest was not different between groups. Respiratory exchange ratio was significantly higher in HE than ME and SC. There were 8 ME and 7 SC with RER's below a physiological range (0.70). This may be a reflection of post-test gas calibrations having not been performed regularly. Barometric pressure changes were not adjusted in the computer software, which may have also accounted for some values below physiological range. Predicted resting energy expenditure (REE) (kcal/day) (Vmax, Sensormedics, Yorba Linda, CA) was significantly lower in ME than SC. However, when measured REE was expressed as kcal/day or kcal/kgFFM/day, no differences were detected.

Table 7 - Resting energy expenditure. [**mean**, ± SD, (range)]

Group	Predicted REE (kcal/day)	VO_2 (l/min)	REE (kcal/day)	REE (kcal/kgFFM/ day)	Respiratory exchange ratio
HE (n = 16)	**1772** ±114 (1603 – 1990)	**0.299** ±0.036 (0.217 – 0.351)	**2042** ±264 (1478 – 2424)	**30** ±2 (25 – 34)	**0.75** ±0.05 (0.66 – 0.82)
ME (n = 20)	**1698** ±130 (1460 – 2007)	**0.289** ±0.029 (0.237 – 0.340)	**1926** ±190 (1606 – 2324)	**31** ±2 (28 – 35)	**0.70** ±0.03 (0.65 – 0.79)
SC (n = 16)	**1836** ±132 (1592 – 2066)	**0.289** ±0.031 (0.232 – 0.329)	**1939** ±236 (1511 – 2262)	**30** ±3 (25 – 35)	**0.72** ±0.04 (0.66 – 0.79)
p values	.006365[a]	.570471	.281920	.252420	.003353[b] .010871[c]

Note: a = ME significantly different than SC
b = HE significantly different than ME
c = HE significantly different than SC

6. 4-day energy expenditure and physical activity record

Energy expenditure was assessed over a 4-day period using the TriTrac R3-D™ accelerometer. Physical activity journals were used to record training duration and intensity.

6.1 TriTrac R3-D™ (Table 8)

The TriTrac R3-D™ provided values for REE (prediction equation), energy expenditure from activity as well as total energy expenditure (REE and activity energy). An average of the 4 days was calculated for each component of energy expenditure. Values were reported as both kcal/day as well as kcal/kgFFM/day. Results indicated that HE expended significantly higher total kcal/day than ME or SC. When expressed as kcal/kgFFM/day, HE and ME were significantly higher than SC although there was no difference between HE and ME. Estimation of REE from the TriTrac R3-D™ resulted in a significantly lower REE in ME than SC when expressed as kcal/day. However, estimated REE of the TriTrac R3-D™ (expressed as kcal/kgFFM/day) showed significant differences between both athlete groups and SC. These values were not used to calculate REE for the energy balance equation. Energy expenditure from exercise, using the TriTrac R3-D™, indicated significantly higher activity calories in HE than ME and SC as well as in ME than SC (both kcal/day and kcal/kgFFM/day).

6.2 Self-reported activity logbook (Table 9)

Subjects recorded duration of exercise performed over the 4-day period. Subjects reported perceived exertion for their activities on an intensity scale ranging from 1 to 5. Total time of exercise over the 4-day period was significantly lower in SC than HE and ME with no differences observed between HE and ME. Total time spent at intensity "1" and "2" were not different between groups. Total time spent at intensities of "3" and "4" were significantly higher in HE and ME than SC with no

differences between HE and ME. Total time spent at intensity "5" was significantly different between HE and SC.

7. Eating attitudes

Eating attitudes were assessed using the EDI and DEBQ-R questionnaires. Scores for each EDI subscale and DEBQ-R are shown in Table 10. There were no significant differences between any of the groups.

Table 8 - Estimated energy expenditure assessment over 4-day period using TriTrac R3-D™. [**mean**, ± SD, (range)]

Group	Total (kcal/day)	REE (kcal/day)	Activity (kcal/day)	Total (kcal/kgFFM/day)	REE (kcal/kgFFM/day)	Activity (kcal/kgFFM/day)
HE (n = 16)	**2977** ±348 (2435 – 3815)	**2027** ±150 (1800 – 2318)	**951** ±245 (578 – 1496)	**44** ±3.3 (38 - 48)	**30** ±1.4 (27 - 32)	**14** ±3.0 (9 - 17)
ME (n = 20)	**2616** ±212 (2298 – 2949)	**1921** ±166 (1684 – 2340)	**695** ±150 (441 – 977)	**42** ±3.9 (37 - 50)	**31** ±1.6 (28 - 35)	**11** ±3.0 (8 - 17)
SC (n = 15)*	**2510** ±206 (2175 – 2928)	**2097** ±181 (1771 – 2390)	**413** ±127 (208 – 667)	**38** ±2.9 (34 - 47)	**32** ±1.8 (28 - 35)	**6** ±2.1 (3 - 12)
p value	.000316[b] .000135[c]	.009644[a]	.000266[b] .000126[c] .000153[a]	.004340[a] .000196[c]	.001668[c] .024671[a]	.000120[a] .000126[c] .006367[b]

Note: a = ME significantly different than SC
b = HE significantly different than ME
c = HE significantly different than SC

*SC = Missing data from one subject in SC group. Sample size for SC = 15.

Table 9 - Duration of self-reported activity over 4-day period. [**mean**, ± SD, (range)]

Group	Total time exercising (min)	Intensity "1" (min)	Intensity "2" (min)	Intensity "3" (min)	Intensity "4" (min)	Intensity "5" (min)
HE (n = 16)	**442** ±184 (154 – 764)	**101** ±66 (0 – 331)	**73** ±59 (0 – 170)	**176** ±150 (0 – 425)	**89** ±67 (0 – 190)	**38** ±59 (0 – 185)
ME (n = 20)	**340** ±126 (169 – 582)	**87** ±33 (0 – 390)	**55** ±72 (0 – 230)	**177** ±97 (0 – 408)	**116** ±91 (0 – 343)	**20** ±35 (0 – 120)
SC (n = 16)	**129** ±118 (0 – 315)	**42** ±22 (0 – 120)	**32** ±61 (0 – 180)	**49** ±79 (0 - 300)	**26** ±46 (0 – 175)	**0** (0)
p values	.000126[c] .000118[a]	.284394	.203314	.001854[c] .004676[a]	.013438[c] .001905[a]	.018831[c]

Note: a = ME significantly different than SC
c = HE significantly different than SC

Table 10 - EDI subscale and DEBQ-R scores. [**mean**, ± SD, (range)]

Group	EDI Thinness	EDI Awareness	EDI Bulimia	EDI Dissatis-faction	EDI Ineffect-iveness	EDI Maturity	EDI Perfection-ism	EDI Distrust	DEBQ-Restraint
HE (n = 15)*	**1.1** ±1.7 (0 –3)	**0.8** ±1.8 (0 – 7)	**0.7** ±1.5 (0 – 5)	**0.6** ±1.1 (0 – 3)	**0.2** ±0.6 (0 – 2)	**2.3** ±1.9 (0 – 6)	**3.4** ±2.7 (0 – 8)	**0.8** ±1.6 (0 – 5)	**18.2** ±6.1 (10 – 30)
ME (n = 20)	**0.6** ±1.7 (0 – 7)	**0.5** ±0.6 (0 – 2)	**0.2** ±0.5 (0 – 2)	**1.6** ±3.0 (0 – 11)	**0.6** ±0.9 (0 – 3)	**1.4** ±2.0 (0 – 6)	**4.1** ±3.2 (0 – 10)	**2.4** ±3.0 (0 – 11)	**17.3** ±4.8 (12 – 32)
SC (n = 16)	**0.8** ±1.0 (0 – 3)	**0.8** ±1.7 (0 – 5)	**0.3** ±0.9 (0 – 3)	**1.6** ±2.4 (0 – 8)	**1.8** ±4.0 (0 – 16)	**3.0** ±3.2 (0 – 12)	**4.1** ±3.9 (0 – 15)	**2.2** ±2.4 (0 – 8)	**19.1** ±6.8 (10 – 32)
p value	.598764	.756743	.225871	.410625	.133182	.128830	.797871	.150901	.652024

Note: *HE = Missing data from one subject in HE group. Sample size for HE = 15.

8. Energy balance (Table 11)

Energy balance was calculated using the following variables: [measured REE (Vmax) (kcal/day) + TEF (10% of REE) (kcal/day) + activity calories from TriTrac R3-D™ (kcal/day)] subtracted from average total daily energy intake (kcal/day). A negative value reflects negative energy balance where a positive value reflects adequate energy intake for energy expenditure. When expressed as kcal/day, there were no significant differences between the three groups when Post Hoc analysis was performed (Neuman-Keuls). Energy balance (kcal/kgFFM/day) was significantly higher in ME than SC. Potential errors in REE (some subjects with RER's below 0.70) may account for inaccurate estimations of energy balance.

Table 11 - Energy balance* [**mean**, ± SD, (range)]

Group	Energy Balance (kcal/day)	Energy Balance (kcal/kgFFM/day)
HE (n = 16)	**273** ±543 (-373 – 1252)	**4.4** ±8.9 (-6 – 21.5)
ME (n = 20)	**631** ±523 (-378 – 1735)	**10.2** ±8.4 (-6 – 28.7)
SC (n = 15)**	**190** ±576 (-691 – 1091)	**3.0** ±8.7 (-10.8 – 16.7)
p values	.043810***	.045887[a]

Note: a = ME significantly different than SC

* Energy Balance = Average energy intake (kcal/day) – [measured resting energy expenditure (Vmax) (kcal/day) + thermic effect of feeding (10% of REE) (kcal/day) + energy expenditure from exercise (TriTrac R3-D™) (kcal/day)]

** SC = Missing data from one subject in SC group due to TriTrac R3-D™ data. Sample size for SC = 15.

*** p value after running ANOVA, however Post Hoc analysis (Neuman-Keuls) resulted in no significant differences between groups.

9. Hormonal concentrations (Table 12 and 13)

Total testosterone (Figure 4), free testosterone (Figure 5), prolactin (Figure 6), LH (Figure 7), FSH (Figure 8), total T_4 (Figure 10) and cortisol (Figure 11) were not significantly different between the three groups. Total T_3 (Figure 9) was significantly

lower in the HE than SC. Intra-assay CV's for all hormones were acceptable (range of 2.02 to 5.52).

Table 12 - Total T_3 and Total T_4 concentrations. [**mean**, ± SD, (range)]

Group	Total T_3 (ng/dl)*	Total T_4 (µl/dl)*
HE (n = 16)	**102.0** ±11.4 (79.7 – 120.0)	**6.3** ±0.7 (4.8 – 7.9)
ME (n = 20)	**110.2** ±19.5 (84.7 – 144.2)	**6.8** ±1.1 (5.1 – 8.6)
SC (n = 16)	**117.8** ±16.2 (92.3 – 157.5)	**6.8** ±1.1 (5.3 – 9.6)
p values	.018706[c]	.331258

Note: c = HE significantly different than SC

* SI Units = Conversion Factors

Hormone	Metric Unit	Conversion Factor	SI Unit symbol
Total T_3	ng/dl	0.01536	nmol/l
Total T_4	µl/dl	12.87	nmol/l

Expected values (DPC, 1999)

Hormone	Metric Unit	Median	Central 95% Range
Total T_3	ng/dl	131	86 - 187
Total T_4	µl/dl	n/a**	4.5 - 12.5

** = Not available

Table 13 - Testosterone (free and total), LH, FSH, Prolactin and Cortisol concentrations. [**mean**, ± SD, (range)]

Group	Free Testosterone (pg/ml)*	Total Testosterone (ng/dl)*	LH (mIU/ml)	FSH (mIU/ml)	Prolactin (ng/ml)*	Cortisol (µg/dl)*
HE (n = 16)	**13.6** ±4.3 (8.5 – 24.2)	**411.4** ±127.6 (269.0 – 727.0)	**2.5** ±1.4 (0.5 – 5.6)	**3.2** ±2.1 (0.8 – 9.5)	**8.0** ±3.5 (3.2 – 11.8)	**9.6** ±3.4 (4.1 – 18.4)
ME (n = 20)	**16.1** ±5.2 (6.1 – 26.7)	**434.7** ±153.9 (256.5 – 707.1)	**3.5** ±2.1 (0.7 – 10.8)	**4.0** ±3.1 (0.9 – 13.6)	**7.1** ±2.2 (4.2 – 11.3)	**11.2** ±3.2 (7.1 – 17.4)
SC (n = 16)	**17.5** ±5.7 (7.0 – 30.1)	**432.1** ±131.5 (161.5 – 642.4)	**3.8** ±1.4 (0.7 – 5.5)	**2.6** ±1.5 (1.2 – 6.1)	**6.0** ±1.9 (2.3 – 8.8)	**9.9** ±4.6 (4.3 – 18.8)
p values	.100439	.868588	.191298	.235960	.095491	.400015

Note: a = ME significantly different than SC
b = HE significantly different than ME
c = HE significantly different than SC

* SI Units = Conversion Factors

Hormone	Metric Unit	Conversion Factor	SI Unit symbol
Free Testosterone	pg/ml	3.467	pmol/l
Total Testosterone	ng/dl	0.03467	nmol/l
Prolactin	ng/ml	1.0	µg/l
Cortisol	µg/dl	27.59	nmol/l

Expected values (DPC, 1999)

Hormone	Metric Unit	Median	Central 95% range
Free Testosterone	pg/ml	23	13 -40
Total Testosterone	ng/dl	630	262 - 1836
LH	mIU/ml	2.0	0.4 - 5.7
FSH	mIU/ml	3.9	1.1 - 13.5
Prolactin	ng/ml	4.4	ND** - 15
Cortisol	µg/dl	n/a***	am: 5 - 25; pm: 1/2 of am value

**ND = Not detectable

***n/a - not available

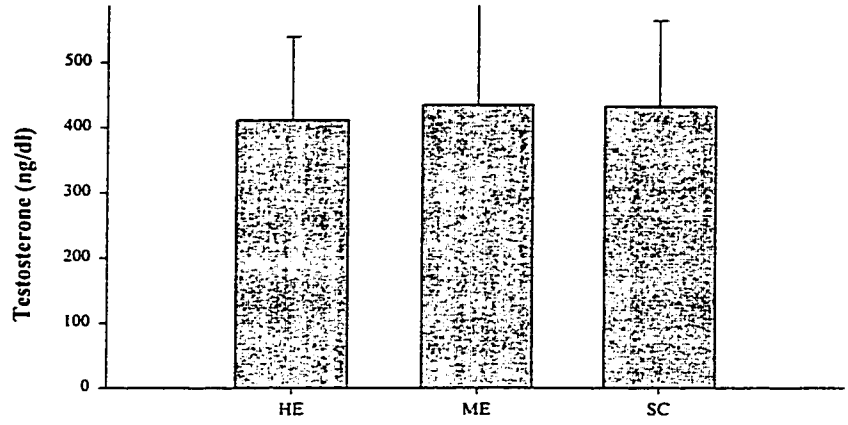


Figure 4 - Serum Testosterone concentration

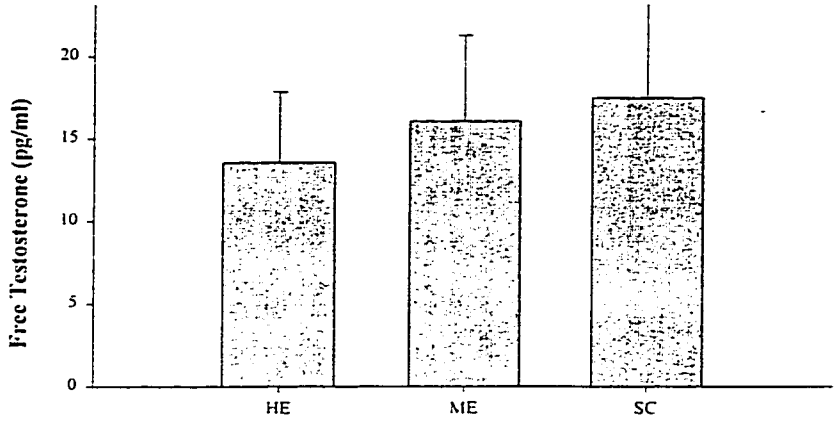


Figure 5 - Serum Free Testosterone concentration

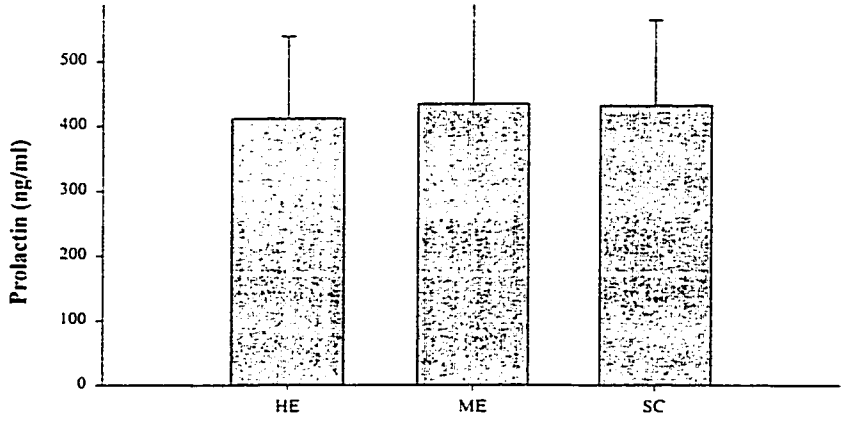


Figure 6 - Serum Prolactin concentration

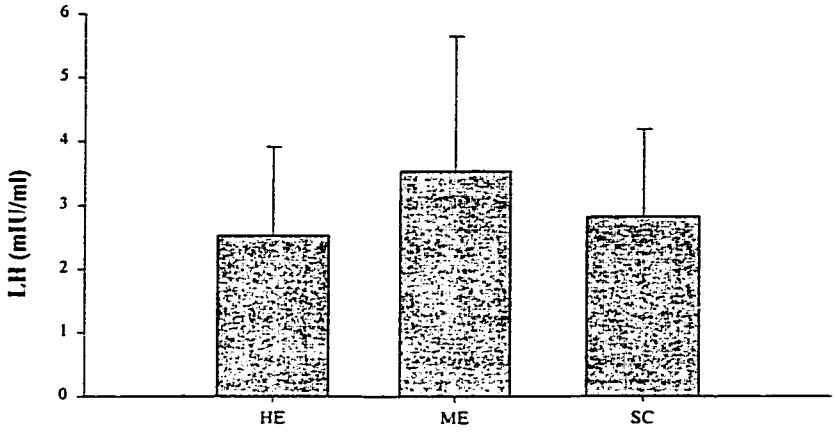


Figure 7 - Serum Luteinizing Hormone concentration

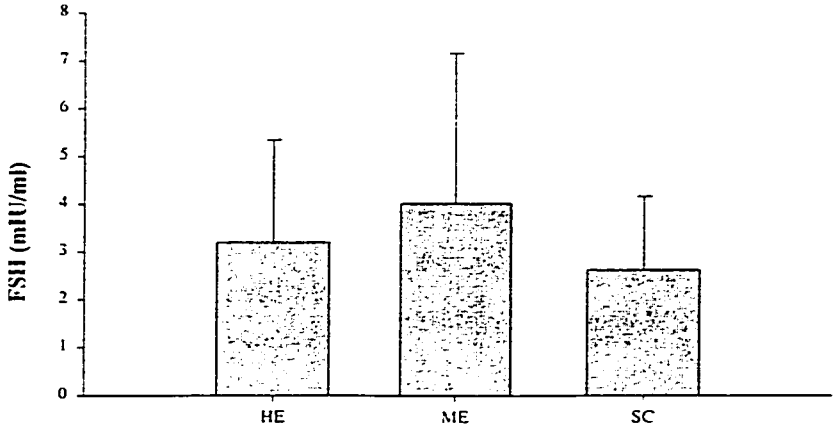
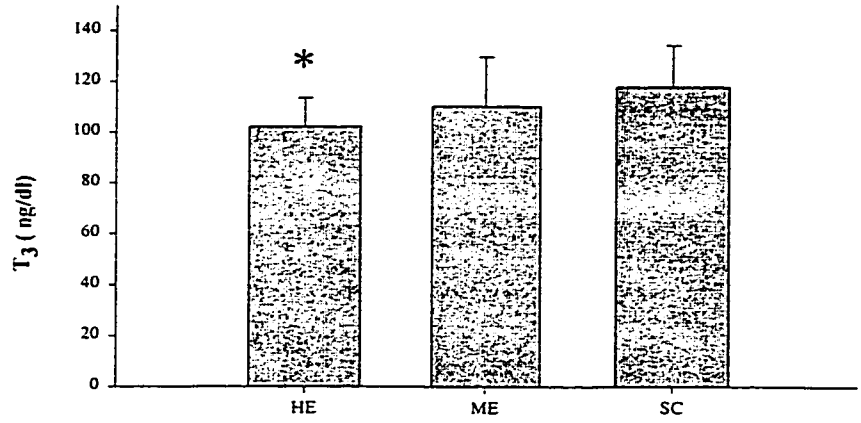


Figure 8 - Serum Follicle Stimulating Hormone concentration



* = HE significantly lower than SC (p = 0.18706)

Figure 9 - Serum T_3 concentration

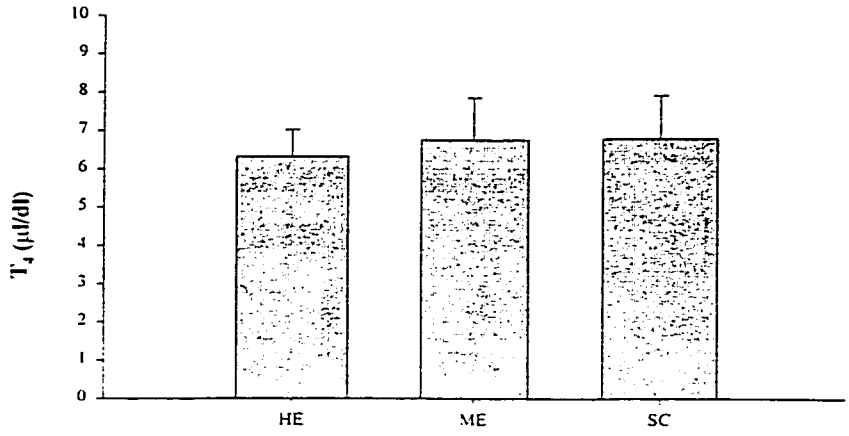


Figure 10 - Serum T_4 concentration

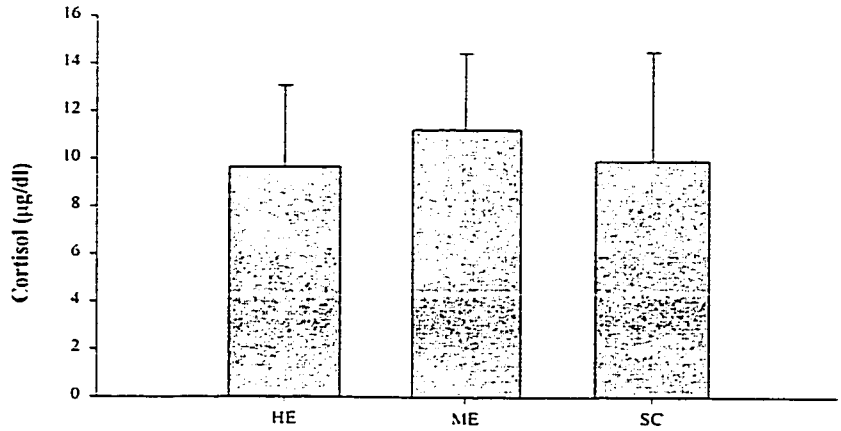


Figure 11 - Serum Cortisol concentration

CHAPTER 5

DISCUSSION

1. Introduction

Total energy expenditure, including REE, TEF, energy from daily activities as well as from exercise, has not been measured in male athletes as it relates to suppressed reproductive hormones. However, volume of training has been shown to correlate with increased disturbances to the reproductive system in male athletes (De Souza et al, 1994a). The relationships of energy intake and macronutrient sources with physiological changes to the reproductive hormone profile have been evaluated in female athletes (Harber et al, 1998; Loucks et al, 1994a; Loucks et al, 1994b; Olson et al, 1995; Williams et al, 1995) and to a lesser extent, in male athletes (Cameron et al, 1991; De Souza et al, 1994a; Rojdmark, 1987). Insufficient energy intake for exercise performed in certain endurance athletes may contribute to the suppressed reproductive hormone profile observed in some male endurance-trained athletes.

This present study intended to examine the components of energy balance independently, and in combination, in highly-trained endurance (HE), moderately-trained endurance (ME) and sedentary (SC) males. The predominant modality of training for the athletes was cycling. These components included REE, estimated TEF (as 10% of REE), energy expenditure from activity and exercise as well as energy intake. In addition, total testosterone, free testosterone, LH, FSH, prolactin, cortisol, T_3, and T_4 were analyzed for each group. A discussion of the results of the

present study will follow. The hypotheses will be reviewed and where literature exists for comparison of the findings of this study, discussion will be included.

2. Subjects

Subjects were carefully screened regarding training parameters prior to being permitted into the study. Sedentary males (SC) reported exercising less than four hours per week over the last year. The researcher inquired about specific aerobic exercise performed regularly by SC to confirm miss-representation of grouping did not occur. Aerobic fitness tests (VO_2max) contributed to verifying grouping. Athletes (ME and HE) were classified as cyclists, as the predominant mode of training was on bicycles. However, within this sample population there was lack of homogeneity as the term "cyclist" included road cyclists, mountain bikers and triathletes. Reported activities in the 4-day logbook reflected a variety of modes of training, including swimming and running. Whereas most research has been focused on runners (Are et al, 1993a; Hackney et al, 1988; Hackney et al, 1990; Jensen et al, 1995; MacConnie et al, 1986; McColl et al, 1989; Wheeler et al, 1984), few have analyzed athletes with reported diversity in training modalities (Lucia et al, 1996; Neary et al, 1994).

The marginal distinction between the athletic groups may have contributed to the lack of differences among HE and ME with respect energy balance measures as well as reproductive hormonal profiles. De Souza et al (1994a) were able to distinguish differences in circulating levels of testosterone between highly-trained runners and moderately-trained runners. Athlete classification based upon mileage

may have been sufficiently different in De Souza's (1994a) study whereas HE and ME may have been too similar. Comparison of mileage is not relevant, however, as the training modes varied. Collapsing athletes together and comparing with SC also resulted in no differences regarding energy balance variables or reproductive hormone profiles.

3. Energy intake

Energy intake was measured using a 4-day dietary analysis tool. It had been hypothesized that there would be no differences in energy intake between the athletes and controls as previous research in females has demonstrated (Harber et al, 1998), however this was not reproduced in this study of male endurance athletes. Average daily energy intake was significantly higher in the HE and ME than SC. There were no differences between HE and ME. Elevated energy intakes are consistent with estimated higher energy expenditure in the active males. In absence of measured energy expenditure one could not indicate whether the ME was ingesting excess energy or conversely, if the HE was ingesting insufficient energy. Further analysis of energy expenditure and state of energy balance is necessary to address this. Several studies have identified that highly trained female athletes report similar energy intake to sedentary females (Loucks et al, 1989; Myerson et al, 1991; Wilmore et al, 1992). This information has not been illustrated in males.

Macronutrient intake was also examined in the present study using the 4-day dietary analysis. No differences in macronutrient intake between groups were detected other than ME having lower fat intake than SC. Previous research has found

lower fat intake in female amenorrheic athletes than eumenorrheic athletes (Harber et al, 1998), as well as increased carbohydrate intake in high-mileage male runners compared to moderate-mileage runners or sedentary controls (De Souza et al, 1994a). HE, ME and SC may have had different dietary choices than De Souza et al's (1994a) groups.

Use of 4-day dietary records for assessing habitual energy and macronutrient intake has well-documented limitations. Subject compliance and accuracy of self-reporting may affect results (Mertz et al, 1991). Further, accurate representation of average diet is limited when assessing 4 days. Longer recording periods and repetitive measurements over time would strengthen certainty of representation of chronic eating habits. Use of the 4-day dietary record may have under-represented, or conversely, over-represented energy intake for the three groups.

In this study, it was hypothesized that aberrant eating attitudes would not be present in any group. The EDI-Subscales and DEBQ-Restraint questionnaires were administered as a process of identifying potential disordered eating attitudes. There were no differences between groups and there were no indications of disordered eating behaviors in any of the groups. This reduced the possibility that differences in hormones measured would be resultant from disordered eating tendencies rather than state of energy balance or its various components. Eating attitudes have been explored in female athletes and there is limited information regarding male athletes. Wheeler et al (1986) compared eating attitudes of high endurance male runners, moderate endurance male runners and sedentary controls. High endurance runners demonstrated increased tendency towards aberrant eating attitudes. Restrained eating

is also associated with decreased energy intake in males (Tepper et al, 1996). Anorexic tendencies have been associated with altered hormones of the reproductive axis in females (Fichter et al, 1989). Similar research has not been done in males. These tendencies were not observed in either HE or ME in this study.

4. Resting energy expenditure

It was hypothesized that REE of HE would be lower than ME or SC. REE obtained from indirect calorimetry did not differ between groups in this study. The measured REE differed from prediction equations. The prediction formula used with the TriTrac R3-D™ (Hemokinetics Inc., 1996) as well as the predicted measure provided by the metabolic cart (Vmax, Sensormedics) for REE resulted in ME having lower REE than SC. Previous research had suggested that high endurance athletes may have lower absolute REE than sedentary controls (Tremblay et al, 1997, Thompson et al, 1995). However, other research has found absolute REE values to be higher in female athletes than non-athletes unless expressed as kcal/kgFFM/day, where there were no differences observed (Wilmore et al, 1992). Further, other research has found no differences in absolute REE between trained athletes and untrained (Burke et al, 1993). As previous literature has reported varied REE values between trained and untrained, results of the present study are not inconsistent. This study suggests that HE and ME do not have different metabolic efficiency in non-activity related energy expenditure as has been previously reported in female athletes (Burke et al, 1993; Myerson et al, 1991). Rather, training status may have had no effect on REE.

Limitations in REE values exited however, due to technical errors in performing the indirect calorimetry. There were 17 of the 52 subjects (representing the three groups) who had RER's below 0.70. This may have indicated some gas calibration errors during the testing. Gas calibrations were performed before and during testing, however post-test gas calibrations were not routinely performed. Recommendations for future testing would include performing both pre- and post-test gas calibrations as well as to ensure correct entry of air temperature and barometric pressure. Implications of the low RER's include less certainty in the accuracy of energy expenditure at rest for the groups. This may potentially impact the energy balance equation. Subject compliance to no exercise for 24 hours prior to testing, 12 hour fast prior to testing as well as arriving to the testing lab in a relaxed state are also inherent limiting factors associated with indirect calorimetry testing for REE. This, too, may have an impact upon the energy balance equation.

Thermic effect of feeding (TEF) was not different between groups as it was calculated as 10% of REE. Previous research on postprandial thermogenesis have shown male athletes to have lower TEF than sedentary controls (Poehlman et al, 1988) while other research has found no differences between athletic and non-athletic individuals (Beidleman et al, 1995; Burke et al, 1993). The previous studies utilized indirect calorimetry measurements following ingestion of a known mixed nutrient composition liquid meal. This method of measuring TEF differed from this study, therefore comparison of results is difficult.

In calculating energy balance, the values obtained from the indirect calorimetry for REE and in turn, TEF, were used. As there were no differences in

expended energy at rest between the three groups, energy expended from exercise and daily activity may be more influential in determining negative energy balance between groups.

5. Exercise energy expenditure

The TriTrac R3-D™ was used as a tool for assessing energy expenditure. It was capable of predicting REE per minute as well as determining energy expenditure from activity per minute (Hemokinetics, Madison, WI, 1996). Energy expenditure from activity, determined from the TriTrac R3-D™, was used in calculating energy balance. The TriTrac R3-D™ does not differentiate between daily activity and exercise therefore these values were combined in the calculation of energy balance. It was hypothesized that energy expenditure from exercise would be higher in HE than ME or SC and higher in ME than SC. In this study, energy expenditure from activity measured from the TriTrac R3-D™ was significantly different, as hypothesized, between all three groups. This was an expected finding as it confirmed that the three groups were training at different durations. 4-day measurements of total energy expenditure (TEE) resulted in HE expending greater kcal/day than ME or SC, however when expressed as kcal/kgFFM/day, both HE and ME were higher than SC with no significant differences between HE and ME. Previous studies on energy expenditure in male athletes have not used the TriTrac R3-D™ as an analytical tool therefore there are no direct comparisons of the energy expenditure findings of this study with others. The TriTrac R3-D™ has not been validate for use in measuring energy expenditure for cycling activity. This may be limiting in interpreting energy

expenditure from activity. Swimming was also not measured by the TriTrac R3-D™. It is possible that there was an under-estimation of energy expenditure from exercise with HE and ME who participated in these two activities frequently.

A second measure of energy expenditure from exercise was obtained from a self-reported activity logbook. Subjects recorded all exercises performed over the 4-day period and indicated the perceived intensities of each activity. Previous research has distinguished HE and ME male runners through duration and mileage (De Souza et al, 1994a). As the athletes in this group were cyclists, it was expected to see time spent exercising to be greater in this population than previously observed durations of highly trained and moderately trained runners. It was also hypothesized that there would be significant differences of time spent at higher intensities of exercise ("3" to "5") in HE than ME or SC and ME than SC. Total time of exercise was significantly higher in HE and ME than SC with no differences between HE and ME. This did not support the hypothesis, however when distribution of intensities at which duration was spent was analyzed HE was significantly higher at "3" and "4" than ME or SC. HE was also significantly higher duration of time at an intensity of "5" than SC. This confirms that intensity of training as well as duration of training was significantly different between the three groups. This is congruent with previous research, which identified suppressed reproductive hormone profiles when differentiating athletic populations by volume/duration of training (De Souza et al, 1994a; Hackney et al, 1990; MacConnie et al, 1986; Wheeler et al, 1984).

6. Energy balance

It was hypothesized that an energy balance equation from the aforementioned components would result in HE being significantly lower (negative energy balance) than ME or SC. In this study, there were no differences between the three groups when expressed as kcal/day. However, when fat free mass was accounted for (kcal/kgFFM/day), ME was significantly higher than SC. There were no other differences observed. This was an unexpected finding. Reviewing the components of energy balance assists in understanding these results. Energy intake was higher in HE and ME compared to SC. On the expenditure portion of the equation, REE was similar between all groups. TEF was therefore similar between groups. Exercise energy expenditure obtained from the TriTrac R3-D™ was significantly higher in HE than ME or SC and higher in ME than SC. Therefore, the higher value for energy balance of ME is likely explained by the similar energy intakes (kcal/day) of ME to HE. It is possible that energy intake of HE and SC were sufficient for exercise performed where for ME, it was may have been excessive intake for expenditure. Conversely, the similar energy balance between HE and SC may indicate that both groups were in a lower state of energy balance, while ME was taking in sufficient energy for exercise performed. As this is the first research to develop an energy balance equation with these measurements, it is not possible to compare with other literature. Therefore, it is not known what absolute value indicates sufficient energy balance for the maintenance of all basic life functions, including reproductive function. Beidleman et al (1995) measured energy balance in female runners and sedentary controls. Methodological differences of calculating energy balance (energy

expenditure from exercise and TEF) may account for differences in results as their findings indicated an energy deficit (negative energy balance) in both populations, with no significant differences between groups unlike the positive energy balances among the three groups in this study. It is also possible that technical error previously described in measuring REE may have affect energy balance results for HE, ME and SC as values for REE and TEF may be inaccurate for some subjects.

7. Hormone concentrations

It was hypothesized that total and free testosterone would be significantly lower in HE than ME or SC. There were no significant differences between groups for either of these hormones. It was also hypothesized that LH, FSH and prolactin would be lower in HE than ME or SC. There were also no significant differences found between the three groups with respect to these hormones. Cortisol was hypothesized to be higher in the HE group than ME or SC and there were no differences observed between groups. These hormones fell within normal ranges for healthy males.

Training may not have been sufficiently different between HE and ME to elicit differences. Conversely, a positive energy balance for HE may eliminated energetic challenges, thus reproductive hormone profiles were not different than ME or SC. These results differ from previous research. There is sufficient literature indicating there to be lower total testosterone in the highly trained than sedentary males (Arce et al, 1993b; De Souza et al, 1994a; Hackney et al, 1988; Hackney et al, 1990; Wheeler et al, 1984). An intervention of increase in training has also resulted

in a decrease in total testosterone (Griffith et al, 1990; Roberts et al, 1993; Wheeler et al, 1991). Further, distinction of weekly mileage or duration of running has been proposed as a potential variable for observed decreases in testosterone between highly trained and moderately trained male athletes (De Souza et al, 1994a). Along the H-P-G axis, there have also been noted decreases in LH (McColl et al, 1989) and prolactin (Wheeler et al, 1991; Wheeler et al, 1984) when comparing highly trained to sedentary males or the influence of increased training in previously sedentary males. However, there are also several studies that have found no differences in production of hormones along the H-P-G axis between highly trained and sedentary males or longitudinal studies of increased training in males. Jensen et al (1995) found no differences in total testosterone, LH, FSH or prolactin with increased training, although lack of a control group raises concerns with these findings (Miller et al, 1997). Hackney et al (1990) found no differences in free testosterone, LH or prolactin between trained and untrained males. Although modes of measuring LH and FSH, including single and multiple interval samples, have been used, other studies have also found no differences in these hormones between groups of athletic and non-athletic males (Arce et al, 1993b; De Souza et al, 1994a; Hackney et al, 1988; Wheeler et al, 1984; Wheeler et al, 1991). Similarly, the results of prolactin between these populations have been controversial in that some studies have also found no differences (Arce et al, 1993b; De Souza et al, 1994a; Hackney et al, 1988). Cortisol has not been reviewed to the same extent in high endurance males compared with moderate endurance or sedentary males in relation to energy expenditure and intake. Training intervention, with increased mileage, resulted in increased cortisol

levels (Roberts et al, 1993) and highly trained endurance athletes had elevated cortisol levels at rest compared to moderately-trained athletes or sedentary controls (Luger et al, 1987). Competitive cyclists had higher cortisol than sedentary controls and had increases in cortisol following a 7-week training intervention (Neary et al, 1994).

Thyroid hormones were analyzed in this present study as well. It was hypothesized that T_3 and T_4 would be significantly lower in HE than ME or SC. There were no differences observed between groups with respect to T_4, but HE had significantly lower T_3 than SC. T_3 for ME was not different compared with other groups. Lower T_3 in HE may be a marker of energy insufficiency. Although T_3 was lower in HE in this study, values fell within normal healthy range for males. Amenorrheic athletes have displayed lower T_3 than eumenorrheic athletes or sedentary females (Harber et al 1998). Similarly, reduced energy availability has resulted in decreased T_3 in females (Loucks et al, 1994b). There has been limited research on T_3 and reproductive function in females and there is no data presently available in males.

8. General discussion

The lower T_3 observed in HE in this study may provide further insight as to the role of exercise and energy balance on reproductive function in males. Although there were no differences in the hormones of the H-P-G axis (testosterone, LH, FSH or prolactin), a marker of insufficient energy (lower T_3) may have indicated a disruption to energy balance. As previously mentioned, it is not know at present what

values in an energy balance equation are considered as insufficient energy to maintain basic life function, including reproductive function. However, ME had significantly higher energy balance values than SC (kcal/kgFFM/day), where there were no differences in energy balance values between HE and SC. There were no differences in T_3 between ME and SC or ME and HE, yet T_3 was lower in HE than SC. In this study, HE may have displayed a marker of insufficient energy with significantly lower T_3 however, the data does not support that energy balance values were different between HE and SC. As has been previously discussed, there are inherent margins of error associated with measuring energy intake and energy expenditure (resting and from exercise). As some subjects' RER values were below physiological ranges during the REE measure, the values may not be reliable. This would discount the value of the REE contribution of the energy balance equation, thus miss-representing energy balance for those individuals. Conversely, it is possible that serum samples of T_3 are more sensitive measurements of energy insufficiency than the components of the energy balance equation. This may explain why T_3 was significantly lower in HE than SC, yet energy balance was not different. On the other hand, the difference in T_3 could be a result of a single serum sample. Had there been an observed trend in energy balance values and T_3, greater understanding of the implications of lower T_3 and, or energy balance values would exist. Currently, this hypothesis requires further investigation.

The research of De Souza et al (1994a) is very comprehensive in that it compares many hormones as well as sperm quality of male runners with sedentary males. Where differences were found in testosterone between high mileage and

moderate mileage runners, the possible variable analyzed by the researchers was volume of training. Although energy intake was evaluated, REE was not included nor was energy balance assessed. This present study was not able to reproduce the same hormonal differences observed in De Souza et al (1994a), although a more thorough analysis of variables was included. As there were no observed differences in hormones of the testes or the anterior pituitary between the three groups, the roles of energy intake, REE, TEF, exercise energy expenditure and energy balance in the maintenance of the H-P-G axis remain uncertain.

A positive relationship between duration and, or mileage of running and disruption to the H-P-G axis has been established in previous research (De Souza et al, 1994a). HE athletes in this study did not present with different hormonal profiles of the H-P-G than ME or SC, despite having different energy expenditures (TriTrac R3-D™). However, as total duration of exercise (logbook) did not differ between HE and ME, there may not have been sufficient differentiation in training classifications. Conversely, it may also be suggested that all three groups, regardless of energy expenditure, ingested sufficient energy intake. This can be reflected in the higher energy intake in HE and ME than SC. Energy balance values were all positive for the three groups, therefore appeared to be sufficient for maintenance of basic life functions for all groups. Previous research that has identified alterations to reproductive function in highly trained athletes has failed to analyze energy balance. Further investigation of such populations may indicate that those athletes with lower total and free testosterone, LH, FSH, and prolactin may have higher energy expenditure and may also have negative energy balance.

There are several differences between this present study and previous research that may partially account for the unique findings of this study. Running has been the primary mode of training that has been analyzed in comparing reproductive function of male athletes and non-athletes (Are et al, 1993a; Hackney et al, 1988; Hackney et al, 1990; Jensen et al, 1995; MacConnie et al, 1986; McColl et al, 1989; Wheeler et al, 1984). Where mileage and duration of running have been correlated with lower circulating testosterone in males (De Souza et al, 1994a), this has not been explored with cyclists. Volume threshold identification (De Souza et al, 1994a) was considered when identifying group differentiation through duration of training per week in this study. It is possible that the duration of exercise chosen for differentiation of groups (training classification) in this study was not sufficient to elicit changes in hormonal profiles of the H-P-G axis between HE and ME or SC. The mechanical efficiency of cycling (Farcia, 1992) compared to running may also influence energy expenditure of exercise, thus impacting the energy balance equation. This may partially explain differences in hormonal profiles between this study and De Souza et al (1994a) as higher volumes of cycling than what was observed in this present study may be needed to elicit energy expenditure of highly-trained runners. Where energy balance and, or energy expenditure may be variables influencing various hormones, comparing differences in energy expenditure between sports may be difficult.

This study focused on male cyclists. This categorization of athletes included competitive and recreational road cyclist, mountain bikers and triathletes. As some of the males in HE and SC were not pure cyclists (performed cross-training activities

such as running, swimming), the lack of homogeneity in training regiments may have contributed to the findings. De Souza et al (1994a) did not report having diversity in training between subjects.

The self-recording of physical activity in the logbooks differed between SC and the athletes. SC tended to record daily active living activities ("walk to school", "shovel snow") amongst physical activities while the athletes strictly reported exercise bouts. This reflects differences between groups of perception on physical activity. This discrepancy may have affected energy expenditure expressed as "duration of self-reported activity" in that SC may have been over-represented or ME and HE may have been under-represented.

Another variable that is unique to this study is the use of an accelerometer for measuring energy expenditure. The TriTrac R3-D™, which was used in this study, is able to measure movement of activity and calculate energy expenditure from this. However, it does not calculate the intensity of exercise when cycling. No other data using the TriTrac R3-D™ as a measure of energy expenditure when cycling are available. As well, there are currently no other data supporting the use of the TriTrac R3-D™ in analyzing energy expenditure as it relates to changes within the male reproductive system. Further, eleven subjects participated in swimming as part of their training or leisure and were unable to wear the TriTrac R3-D™ while exercising in the water. SC spent a total of 115 minutes swimming (among 3 subjects), ME spent 135 minutes swimming (among 2 people), while HE spent 625 minutes (among 6 subjects) in the water. These data could not be entered into the calculation of energy balance. Where the tool is informative regarding daily active living energy

expenditure, it may be limiting in its application to all sports. This could impact the calculation of total energy expenditure as well as energy balance. It should be noted, however, that analysis of energy expenditure from the TriTrac R3-D™ resulted in significant differences between all three groups. Whether the differences would have been larger with a more precise measurement of energy expenditure remains to be determined.

Subject compliance in wearing the TriTrac R3-D™ at all times (other than sleep or involvement in water activities) as well as subject compliance and accuracy in recording activities in logbooks (durations and intensities) and in recording dietary analysis were recognized limitations to the study. All activities were analyzed with respect to intensity and duration of training, however these values were not utilized in the energy balance equation. Several complications related to the TriTrac R3-D™ arose that may have also slightly affected expected results. The accelerometer failed on five occasions and these subjects were asked to repeat their 4-day analysis. Data were lost for one subject in the SC group with respect to TriTrac R3-D™ information. Ensuring subjects maintained their regular exercise and diet regimens during the 4-day analysis and that this was reflective of weekly lifestyle were also potential limitations. Under-representation of exercise in HE or conversely, over-representation of exercise or diet in ME would affect energy balance calculations as well as the various components of the energy balance equation.

To further investigate the characteristics of male athletes, HE and ME were collapsed into one group and compared with SC (Table 14). As expected, the athletes had significantly higher scores in total time exercising (min) and energy expenditure

from exercise (TriTrac R3-D™) as well as total energy expenditure (TriTrac R3-D™). REE was not different between the two groups and energy intake was significantly higher in the athletes. Energy balance values did not differ significantly between athletes and non-athletes. In analyzing the hormones, T_3 remained significantly lower in the athletes than the non-athletes. The previous profiles of the other hormones remained unchanged when the athletes were combined.

Table 14 – Comparison of various measures between athletes and non-athletes. [**mean**]

Group	VO_2max (ml/kg/min)	Total time exercising ** (min)	REE (kcal/day)	TriTrac R-3D™ "activity" (kcal/day)	Energy Intake (kcal/day)	Energy Balance (kcal/day)	T_3 (ng/dl)
Athletes (n = 36)***	**60.5**	**418**	**1978**	**809**	**3456**	**472**	**106.6**
SC (n = 16)	**42.6**	**129**	**1939**	**413**	**2709**	**190**	**117.8**
p value	.000115*	.000115*	.582839	.000117*	.000124*	.107812	.028758*

Note: * = Athletes significantly different than SC
** = Average of 4-day self-reported activity logbook
*** = n = 35 for TriTrac R-3D™ "activity" as well as for Energy Balance.

9. Summary

Although previous research has indicated that male endurance runners have significantly different hormonal profiles of the H-P-G than sedentary males (Arce et al, 1993b; De Souza et al, 1994a; Hackney et al, 1988; Hackney et al, 1990; Wheeler et al, 1984), similar findings were not reproduced when high-endurance, moderate-endurance cyclists and sedentary males were evaluated in this study. A marker of insufficient energy (low T_3) was identified in HE male, however there were no further indications of suppressed reproductive hormones in this sample population. Energy expenditure was higher in athletes than non-athletes. Energy intake was not different between HE and ME, although both were higher than SC. REE did not differ

between groups. Calculated energy balance resulted in no differences between groups, unless expressed as kcal/kgFFM/day, where there were no differences between HE and ME or SC yet energy balance for ME was significantly higher than SC.

The results of this study suggest that highly trained endurance males athletes may not have different hormonal profiles than moderately trained endurance males or sedentary males. These highly trained athletes may be ingesting sufficient energy for energy expenditure, thus resulting in a positive energy balance and therefore no disruption to the H-PG axis. Other research, which has identified differences in hormonal profiles of highly trained, moderately trained and non-athletic populations have not analyzed energy balance (Arce et al. 1993b; De Souza et al, 1994a; Hackney et al, 1988; Hackney et al, 1990; McColl et al, 1989; Wheeler et al, 1984). However, decreased T_3 in HE compared to SC may indicate insufficient energy balance which may not be detectable with the current method of calculating energy balance. Methodological limitations may also contribute to the uncertainty of state of energy balance in HE male cyclists.

Further investigation into energy balance and its various components as each relates the male reproductive system, both hormones and spermatogenesis, in male athletes is merited. This study provided insight into methods of calculating energy balance including the usage of the TriTrac R3-D™. This study also provided information regarding male cyclists whereas research to date has focused predominantly on runners when assessing the effects of exercise on male reproductive hormones.

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APPENDIX A

Ethical Approval

Ethics Review Approval

The Ethics Committee of the Faculty of Physical Education and Recreation (University of Alberta):

Name	Position
Dr. Jane Watkinson	Professor and Associate Dean
Dr. Dick Jones	Professor, Pulmonary Medicine

have reviewed the proposal entitled:

Dietary intake, resting metabolic rate and activity levels in sedentary and athletic men
submitted by

Vicki Harber, Kelly Mackenzie

X finds it within acceptable standards for human experimentation

_____ finds it within acceptable standards subject to the following revisions:

_____ finds it unacceptable in its present form

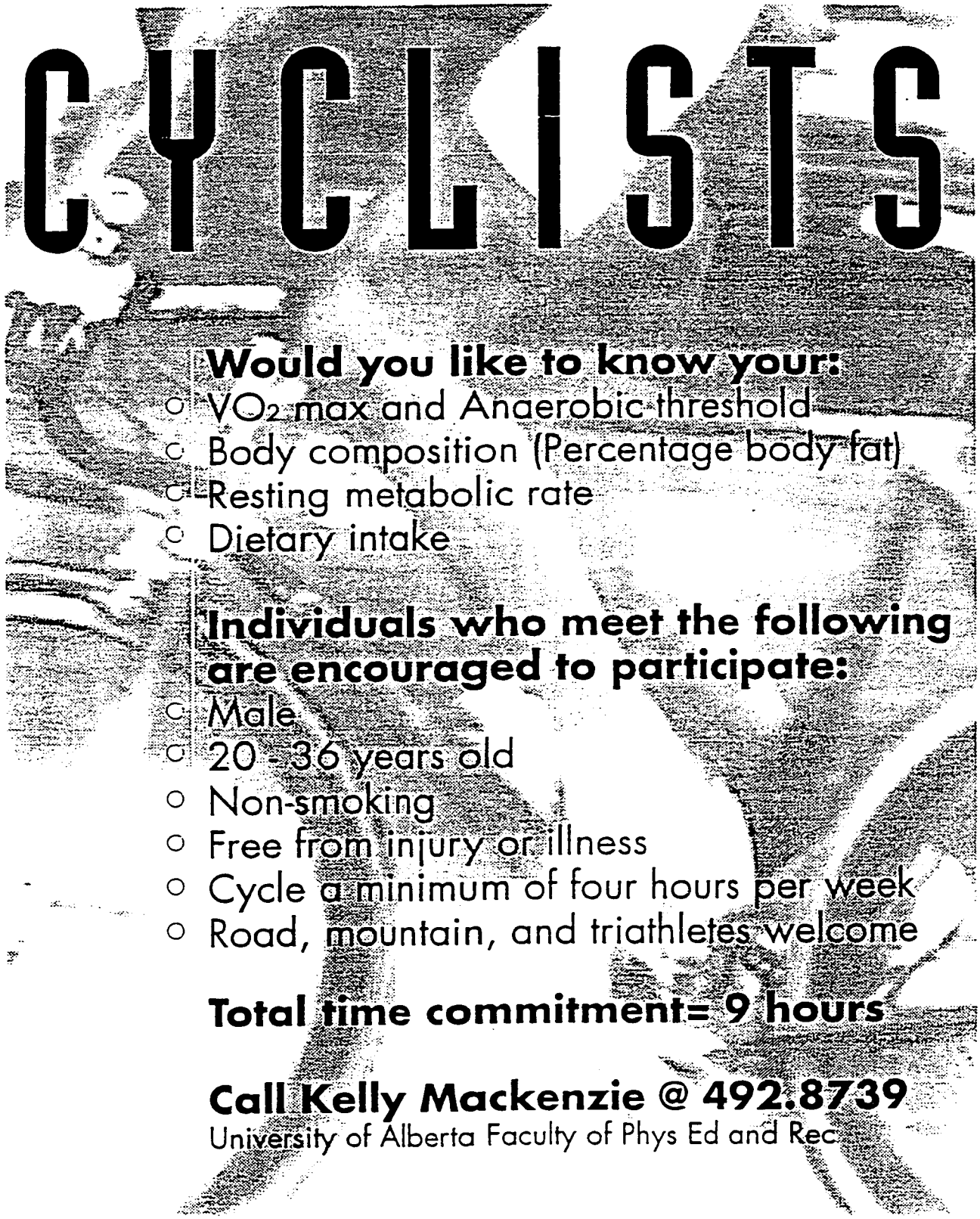
Jane Watkinson

Dr. Jane Watkinson, Chair
Faculty of Physical Education and Recreation

June 5, 1998
Date

APPENDIX B

Recruitment Posters


CYCLISTS
Would you like to know your:
VO2 max and Anaerobic threshold
Body composition (Percentage body fat)
Resting metabolic rate
Dietary intake
Individuals who meet the following are encouraged to participate:
Male
20 - 36 years old
Non-smoking
Free from injury or illness
Cycle a minimum of four hours per week
Road, mountain, and triathletes welcome
Total time commitment= 9 hours
Call Kelly Mackenzie @ 492.8739
University of Alberta Faculty of Phys Ed and Rec

Volunteers Needed

Would you like to know your:

- Aerobic Fitness
- Percentage body fat
- Resting metabolic rate
- Dietary intake

Individuals who meet the following are encouraged to participate:

- Male
- 20 - 36 years old
- Non-smoking
- Free from injury or illness
- Exercise less than two hours per week

Total time commitment= 9 hours

Call Kelly Mackenzie @ 492.8739
University of Alberta Faculty of Phys Ed and Rec

APPENDIX C

Informed Consent

UNIVERSITY OF ALBERTA
DEPARTMENT OF PHYSICAL EDUCATION AND RECREATION

Dietary intake, resting metabolic rate and activity levels in sedentary and athletic men.

Investigators: Kelly Mackenzie, Dr. Vicki Harber,
Dr. Gordon Bell, Dr. David Cumming

SUBJECT INFORMATION SHEET

PURPOSE:

Endurance exercise may have disruptive effects on the male reproductive hormonal axis. Testosterone levels in male endurance athletes may fall below those of sedentary controls. Within the female athlete population, similar disruptions to the reproductive axis have been noted. Increased energy expenditure through exercise as well as decreased caloric intake have been connected to menstrual dysfunction. An imbalance of energy (an energy gap) in the female athlete may interrupt the processes of the reproductive system as a means of conserving energy. Although reproductive production may have observable clinical consequences in women, disruption to the male reproductive system may be undetected by male athletes. This study intends to examine the relationship between a potential energy gap (energy intake - energy expenditure) and circulating hormone levels in men of varying levels of training.

STUDY PROTOCOL:

Sixty healthy males between the ages of 20 to 36, with no history of disorders or use of medications or ergogenic supplements known to affect hormonal status, will be recruited for this study and asked to complete the following:

Subject Participation will include:

- **Health Status Questionnaire:** Subjects will complete a questionnaire to establish medical history, use of medications, reproductive profile, physical activity history as well as other potential complications. **(Time = 1 hour)**

- **Aerobic Fitness Assessment:** Maximal aerobic consumption (VO_2max) will be determined through progressive exercise to volitional fatigue on a cycle ergometer. Muscle discomfort/soreness, shortness of breath, and abnormal heartbeat and blood pressure are possible side effects associated with maximal aerobic consumption (VO_2max) but are rare in healthy young individuals. **(Time = ½ hour)**

(please continue to next page)

Study Protocol . . .

- **Body Composition:** Sum of five skinfolds will be measured with skin calipers (biceps, triceps, subscapular, supra iliac, and calf muscle sites) and body density will be measured by underwater weighing at the University of Alberta. A swimsuit is worn and the water is pleasantly warm. Changing facilities are located adjacent to the test pool. **(Time = ¾ hour)**

- **Blood Sample:** A single blood sample (20 ml) will be taken between 4 and 6 pm by a physician, registered nurse, or trained personnel. Exercise will not be permitted until after the blood sample has been drawn (daytime workout will not be permitted on blood sample day). Bruising and a small risk of infection are possible (but rare) side effects associated with acute venipunctures. **(Time = ½ hour)**

- **Resting Metabolic Rate:** Oxygen consumption at rest will be measured at the University of Alberta. This will be done in the morning, following a day of rest (no physical activity) and 12 hours of fasting. Subject will arrive in a leisurely fashion and will rest for a ½ hour prior to test being performed. The test itself is approximately ¾ hour. The atmosphere is peaceful and relaxing. **(Time = 1½ hours)**

- **Diet Records:** A 4-day diet record will be completed. This will be analyzed on a computer program to determine each subject's usual caloric intakes.**(Time = 2 hours)**

- **Monitoring of Physical Activity:** A physical activity logbook will be completed over 4 days. A Tri-Trac monitor, which measures all movement and estimates caloric expenditure, will be worn for these same 4 days at all times other than sleep and water-related activities (ie. showers, swimming). It is worn like a beeper – a belt will be supplied for comfort. A heart rate monitor will also be worn around the chest with a wrist receiver (watch band) during all bouts of exercise. **(Time = 1 hour)**

- **Eating Attitudes Questionnaires:** Two eating attitude questionnaires will be completed at one of the scheduled visits. **(Time = ½ hour)**

Total time commitment per subject will approximate 9 hours.

CONFIDENTIALITY:

To ensure confidentiality, raw data will be coded and stored in a locked filing cabinet in a locked office to which only the investigators will have access. Normally data is retained for a period of five years post-publication, after which it may be destroyed.

We strongly encourage questions for clarity and understanding of the above outlined experiment. For further information, please feel free to contact Kelly Mackenzie @ 492-8739 or Dr. Vicki Harber @ 492-1023.
e-mail: kam1@gpu.srv.ualberta.ca

UNIVERSITY OF ALBERTA
DEPARTMENT OF PHYSICAL EDUCATION AND RECREATION

Dietary intake, resting metabolic rate and activity levels in sedentary and athletic men.

Investigators: Kelly Mackenzie, Dr. Vicki Harber, Dr. Gordon Bell, Dr. David Cumming

SUBJECT CONSENT FORM

This study has been satisfactorily explained to me by Kelly Mackenzie, Dr. Vicki Harber or their designate. I understand the necessity for the protocol outlines in the Subject Information sheet. I know that I may contact the persons designated on this form at any time if I have further questions. I have been informed of the possible benefits of joining this research study as well as the possible risks and discomforts. I have been assured that the information obtained from my participation in this study may be published in medical reports, but that my personal records will be kept confidential. I understand that I am free to withdraw from this study at any time without prejudice. I understand that I will be promptly informed of any findings, which may develop during the research period that may affect my willingness to continue participating in the study. I understand that I will be given a copy of the Subject Information Sheet and the signed Consent Form to keep.

Subject's Name (print)	Subject's Signature & Date
Witness' Name (print)	Witness' Signature & Date
Investigator's Name (print)	Investigator's Signature & Date

We strongly encourage questions for clarity and understanding of the above outlined experiment. For further information, please feel free to contact Kelly Mackenzie @ 492-8739 or Dr. Vicki Harber @ 492-1023. e-mail: kam1@gpu.srv.ualberta.ca

APPENDIX D

Demographic Questionnaire

DEMOGRAPHIC QUESTIONNAIRE

Name:_____ **Date:**_____

Age:_____ **years**

Date of Birth:_____

Phone #:_____**(daytime)** _____**(evening)**

Mailing Address:_____

Please fill out the following questions as accurately as possible. Please ask for clarification where needed. All information provided is confidential. Thank you!

Section A

1) Over the last 12 months, describe your level of physical activity (aerobic):
   a) Less than 2 hours of exercise per week
   b) Between 2 - 4 hours per week
   c) Between 5 – 7 hours per week
   d) Between 8 - 10 hours per week
   e) Between 11 – 14 hours per week
   f) Over 15 hours per week

2) Do you belong to a sports team/club?
   Yes _____ (please go to question 3)
   No _____ (please go to question 7a)

Questions 3 – 6 need only be filled out if "yes" was answered for Question 2.

3) Name of team/club:_____

4) Sport:_____

5) Number of years competing in the sport:_____years

6) What is the highest level of competition of which you have been involved (please ✓ and indicate year of competition):

City _____
Provincial_____
Varsity_____
National_____
International_____
Other (please specify)_____

Please go to Question 7b. Describe your workouts as well as other activities in which you also participate.

7 a) Do you participate regularly in leisure sports or activities?

Yes _____ (please go to ***Question 7b***)

No _____ (please go to **Section B**)

7 b) Please list and describe these activities:

Activity	Frequency (sessions/week)	Duration (min/session)	Distance (km/wk) (if applicable)	Speed (km/hr) (if applicable)	Intensity *(see below)*
_____					1 2 3 4 5
_____					1 2 3 4 5
_____					1 2 3 4 5
_____					1 2 3 4 5
_____					1 2 3 4 5
_____					1 2 3 4 5
_____					1 2 3 4 5

Intensity Scale:

1 = Not vigorous at all (Very light)
2 = Somewhat vigorous (Light)
3 = Moderately vigorous (Medium)
4 = Vigorous (Heavy)
5 = Extremely Vigorous (Very very heavy)

Section B

8) Have you had any known reproductive problems?

Yes _____(please specify)

No_____

9) Do you have any medical conditions &/or have you been on any medications &/or used any supplements (ie. health food store) in the past 6 months? Please list all:

_____

_____

10) Do you use anabolic steroids?

Yes _____

No _____

11) Do you smoke?

Yes _____

No _____

Thank you for your time. All information will be kept confidential.

APPENDIX E

Diet Record Information Sheet

WHAT DO YOU EAT ???

You will be recording your daily intake of food and fluids for a series of consecutive days. They must include at least *one* weekend day (Saturday or Sunday).

It is imperative that you record EVERYTHING that you eat and drink (water as well!!!). In addition, you must be as ACCURATE as possible when determining the amount (volume or weight) of the food and drink you are recording. It may be difficult for those in residence or for those who are not in complete control of your food intake (preparation, amount, etc.). Use measuring cups/spoons and weigh scales whenever possible.

HINTS FOR RECORDING DIETARY INTAKE

ACCURACY

1. Accurate Measurement Read the weights or volumes of foods or drinks from packages. Example: milk carton, juice box, chocolate bar, potato chips. A "fistful" of meat = 100 gm, "fistful" veggies = 1 cup, 1 cheese single = 1 oz.

2. Method of cooking Indicate how your food was cooked. Example: fried, steamed, baked, broiled etc.

3. "Extras" Don't forget the EXTRAS. Example: ketchup, mustard, mayonnaise, gravy, or butter.

4. Food Types Be specific about TYPES of food/drink. Example: cheddar cheese, 2% milk, margarine or butter. Whenever possible, identify brand names of the foods.

5. Cooked or Dry Measurement Indicate whether the food measurement is "cooked" or "dry". Example: chicken weight before or after cooked.

6. Specific Parts Indicate the exact part of the food you ate or what was removed before eating. Example: chicken (white or dark, bone in or out, skin or skinless), baked potato (skin or skinless), ground beef (lean, extra lean, or regular).

7. Labels Read the nutritional information label from the container (box/can/bag). This will help identify specific brand food nutrients. If you can't find the specific food during your analysis, then you can enter the required data from the label.

BEVERAGES

8. TEA AND COFFEE should be included as well along with the cream, milk and sugar you add.

9. Don't forget WATER.

10. Yes, you do have to record BEER and ALCOHOL as well.....!!

PREPARED OR RESTAURANT MEALS

11. Use PORTION PAKS whenever possible. Example: salad dressing, butter, jams, peanut butter, cheese. It is easier to quantify the volume of these foods...1 portion pak = 1 tablespoon.

12. Fast Foods Include FAST FOOD items by name. Example: McDonald's, Pizza Hut, Wendy's.

13. Recipes Record the amount/volume of ingredients, the number of servings or volume the entire recipe makes and how many servings or what volume you ate.

14. Restaurant Meals When you eat at a restaurant (other than a fast food place, eg. Earl's), record the name of the meal you ate, list the different ingredients on your plate and the quantities of each.

TAKE THE RECORD BOOK WITH YOU AT ALL TIMES.....IT'S EASIER TO RECORD WHAT YOU'RE EATING.

• • • • • • • • • • • • • • • •

		MENU ITEM	UNIT OF MEAS	No. of Units	DESCRIPTION OF MENU ITEM		
		Enter all foods, beverages, etc. consumed as menu items. For every menu item, include any toppings or additives added to the menu item at the time of eating.	Enter the word: "cup" "ounce" "number" "teaspoon" "tablespoon"		Brand	Type of Flavour	Method of Cooking
MORNING MEAL	Menu Item						
	Toppings or Additives						
	Menu Item						
	Toppings or Additives						
	Menu Item						
	Toppings or Additives						
	Menu Item						
	Toppings or Additives						
	Menu Item						
	Toppings or Additives						
	Menu Item						
	Toppings or Additives						
	Mark (X) One Category	Eaten at Your Home					
		Eaten Away From Your Home					
		Did Not Eat					

DAY ONE

APPENDIX F

Physical Activity Logbooks

Cycling Physical Activity Journal

Name: _____ **Dates of Recording:** _____ - _____

Day 1:					*Day 2:*					*Day 3:*					*Day 4:*				
Duration (min)	*Distance (km)*	Average Speed (km/hr)	H R, Monitor Worn?	Intensity	*Duration (min)*	*Distance (km)*	Average Speed (km/hr)	H R Monitor Worn?	Intensity	*Duration (min)*	*Distance (km)*	Average Speed (km/hr)	H. R. Monitor Worn	Intensity	Duration (min)	*Distance* (km)	Average Speed (km/hr)	H. R. Monitor Worn	Intensity
				1 2 3 4 5					1 2 3 4 5					1 2 3 4 5					1 2 3 4 5
				1 2 3 4 5					1 2 3 4 5					1 2 3 4 5					1 2 3 4 5
				1 2 3 4 5					1 2 3 4 5					1 2 3 4 5					1 2 3 4 5

Intensity Scale: 1 = Not vigorous at all (Very Light)
2 = Somewhat vigorous (Light)
3 = Moderately vigorous (Medium)
4 = Vigorous (Heavy)
5 = Extremely Vigorous (Very Very Heavy)

Comments:

Non-Cycling Physical Activity Journal

Name: ____________________ **Dates of Recording:** ______________ - ______________

	Day 1:		*Day 2:*		*Day 3:*		*Day 4:*	
Activity	**Duration (min)**	**Intensity**	**Duration (min)**	**Intensity**	**Duration (min)**	**Intensity**	**Duration (min)**	**Intensity**
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5

Intensity Scale: 1 = Not vigorous at all (Very Light)
2 = Somewhat vigorous (Light)
3 = Moderately vigorous (Medium)
4 = Vigorous (Heavy)
5 = Extremely Vigorous (Very Very Heavy)

Comments:

Physical Activity Journal

Name: ____________________ **Dates of Recording:** ______________ - _____________

	Day 1:		*Day 2:*		*Day 3:*		*Day 4:*	
Activity	**Duration (min)**	**Intensity**	**Duration (min)**	**Intensity**	**Duration (min)**	**Intensity**	**Duration (min)**	**Intensity**
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5
		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5		1 2 3 4 5

Intensity Scale: 1 = Not vigorous at all (Very Light)
2 = Somewhat vigorous (Light)
3 = Moderately vigorous (Medium)
4 = Vigorous (Heavy)
5 = Extremely Vigorous (Very Very Heavy)

Comments:

APPENDIX G

Sample Questions from EDI & DEBQ-R

Sample questions form EDI questionnaire (Garner et al, 1983)

Always	Usually	Often	Sometimes	Rarely	Never	
( )	( )	( )	( )	( )	( )	1. I eat when I am upset.
( )	( )	( )	( )	( )	( )	7. I think about dieting.
( )	( )	( )	( )	( )	( )	19. I feel satisfied with the shape of my body.
( )	( )	( )	( )	( )	( )	38. I think about bingeing (overeating).
( )	( )	( )	( )	( )	( )	42. I feel that I can achieve my standards.
( )	( )	( )	( )	( )	( )	63. I have extremely high goals.

Sample questions from DEBQ-restraint questionnaire (van Strien et al. 1986)

Very Often	Often	Sometimes	Seldom	Never	
( )	( )	( )	( )	( )	1. When you put on weight. do you eat less than you usually do?
( )	( )	( )	( )	( )	5. Do you deliberately eat foods that are slimming?
( )	( )	( )	( )	( )	7. Do you deliberately eat less in order not to become heavier?
( )	( )	( )	( )	( )	10. Do you take into account your weight with what you eat?

APPENDIX H

Daily Energy Intake Analysis

Table 15 – Daily energy intake analysis. (**mean**, ± SD)

Group	Day	kcals	kcal/kgFFM	%CHO	CHO (g)	% PRO	PRO (g)	% FAT	FAT (g)
HE	1	3655 ±754	56 ±14	62 ±10	580 ±189	15 ±3	132 ±24	24 ±9	96 ±38
ME	1	3532 ±959	57 ±15	58 ±8	521 ±179	15 ±3	132 ±37	25 ±6	97 ±35
SC	1	2995 ±890	46 ±16	51 ±12	392 ±135	15 ±3	111 ±32	33 ±11	112 ±55
HE	2	3469 ±832	51 ±13	58 ±11	520 ±185	15 ±4	133 ±37	25 ±10	98 ±39
ME	2	3439 ±694	55±11	58 ±11	498 ±117	14 ±3	120 ±35	26 ±9	103 ±48
SC	2	2677 ±666	41±12	56 ±11	382 ±112	15 ±4	99 ±33	28 ±8	86 ±35
HE	3	3418 ±711	50 ±9	59 ±12	509 ±174	16 ±4	134 ±44	24 ±11	92 ±51
ME	3	3616 ±755	58 ±14	55 ±17	525 ±157	14 ±4	130 ±49	26 ±9	109 ±49
SC	3	2596 ±660	40 ±11	53 ±9	354 ±121	17 ±7	107 ±41	29 ±5	85 ±25
HE	4	3338 ±934	49 ±13	57 ±10	474 ±125	14 ±4	125 ±55	28 ±8	105 ±43
ME	4	3193 ±562	51 ±9	58 ±10	472 ±122	15 ±4	122 ±33	15 ±4	122 ±33
SC	4	2572 ±638	39 ±9	57 ±10	375 ±125	14 ±3	92 ±40	27 ±11	78 ±34

APPENDIX I

Raw Data

Group	Age (years)	Weight (kg)	Height (cm)
HE1	38	81.0	181.8
HE2	25	90.2	183.0
HE3	28	75.3	179.2
HE4	32	72.5	181.0
HE5	22	66.9	175.5
HE6	34	68.7	173.0
HE7	26	88.0	188.0
HE8	23	82.6	183.0
HE9	25	90.4	193.2
HE10	27	77.9	183.0
HE11	37	77.1	185.0
HE12	29	75.8	183.5
HE13	23	68.0	179.2
HE14	28	79.0	182.5
HE15	22	76.8	182.7
HE16	24	66.6	174.4
ME1	22	57.8	164.7
ME2	30	69.0	175.5
ME3	20	75.7	177.8
ME4	26	82.8	181.4
ME5	29	66.5	171.2
ME6	38	72.2	179.1
ME7	32	70.8	176.0
ME8	24	64.3	182.5
ME9	29	84.2	172.5
ME10	24	98.0	179.0
ME11	26	71.3	182.5
ME12	36	64.2	169.0
ME13	25	73.5	173.2
ME14	20	65.8	177.3
ME15	28	55.5	164.2
ME16	23	71.8	177.0
ME17	26	80.2	183.0
ME18	20	64.5	175.5
ME19	27	64.6	174.0
ME20	35	86.1	187.8
SC1	27	90.5	179.0
SC2	29	97.4	185.0
SC3	26	90.0	185.7
SC4	31	80.7	176.1
SC5	26	62.1	164.9
SC6	25	84.4	174.0
SC7	24	77.8	179.0
SC8	33	72.4	181.3
SC9	26	95.1	192.0
SC10	26	77.2	182.0
SC11	31	74.5	177.7
SC12	28	94.7	192.3
SC13	24	82.9	189.4
SC14	24	75.0	176.8
SC15	35	80.0	176.6
SC16	32	85.0	180.2

Group	Biceps (mm)	Triceps (mm)	Subscap(mm)	Iliac (mm)	Calf (mm)	SOS (mm)
HE1	3.10	6.30	8.25	7.85	3.65	29.15
HE2	7.20	13.75	15.60	22.50	6.00	65.05
HE3	4.35	5.65	9.15	19.50	7.40	46.05
HE4	4.45	5.50	14.55	10.65	4.65	39.80
HE5	4.50	6.20	10.95	7.85	5.25	34.75
HE6	3.65	9.40	11.65	11.60	7.20	43.50
HE7	3.30	4.25	10.70	12.90	5.20	36.35
HE8	4.05	7.15	11.40	14.65	8.80	46.05
HE9	5.00	9.00	10.60	10.80	9.80	45.20
HE10	2.95	4.10	7.45	7.85	4.05	26.40
HE11	2.80	5.30	8.15	6.35	4.90	27.50
HE12	3.60	6.10	9.60	10.10	5.90	35.30
HE13	3.65	8.25	7.75	7.40	5.50	32.55
HE14	3.55	6.00	10.60	8.65	4.70	33.50
HE15	4.00	7.60	10.60	8.80	5.90	36.90
HE16	3.80	8.70	7.30	7.75	6.65	34.20
ME1	3.85	8.20	13.10	14.10	6.00	45.25
ME2	3.05	5.95	7.15	4.90	4.30	25.35
ME3	4.30	8.25	10.65	11.65	7.80	42.65
ME4	3.70	4.70	10.40	7.60	6.00	32.40
ME5	3.35	7.15	10.15	9.70	4.25	34.60
ME6	3.15	5.85	10.25	14.45	5.50	39.20
ME7	4.30	8.40	13.85	13.55	6.15	46.25
ME8	2.75	5.30	8.95	8.00	4.15	29.15
ME9	7.00	12.30	16.45	25.40	10.50	71.65
ME10	12.70	19.70	25.85	30.30	20.45	109.00
ME11	3.30	7.05	11.40	11.40	6.45	39.60
ME12	3.60	8.95	13.95	15.30	6.50	48.30
ME13	4.10	9.80	12.00	13.05	7.85	46.80
ME14	3.40	4.70	7.65	5.75	3.40	24.90
ME15	3.45	7.65	13.50	6.75	4.60	35.95
ME16	4.60	7.00	11.30	8.65	6.10	37.65
ME17	3.30	4.95	9.50	8.15	5.70	31.60
ME18	3.60	6.30	8.45	9.40	6.00	33.75
ME19	3.80	8.60	15.25	16.40	4.70	48.75
ME20	3.00	4.25	8.85	8.40	8.75	33.25
SC1	5.70	11.30	13.70	24.15	15.30	85.65
SC2	4.55	9.45	14.95	29.45	9.90	66.30
SC3	7.50	8.60	19.85	18.05	6.50	52.30
SC4	5.80	8.65	14.05	24.35	8.15	69.50
SC5	6.75	9.40	17.65	24.00	7.85	65.65
SC6	9.05	9.30	16.40	20.10	6.80	68.95
SC7	4.20	11.50	8.50	19.10	5.10	48.40
SC8	5.00	13.70	9.30	24.55	9.30	68.85
SC9	5.70	15.20	22.60	27.30	12.30	83.60
SC10	3.90	12.20	19.45	16.80	10.60	62.45
SC11	5.20	12.55	25.20	24.45	9.15	74.55
SC12	5.80	8.95	12.95	21.95	5.40	55.05
SC13	5.90	14.10	15.50	20.15	10.00	60.95
SC14	8.10	9.60	15.45	24.00	5.95	59.10
SC15	6.60	15.05	26.20	22.70	12.90	79.45
SC16	4.70	14.95	20.25	14.95	10.90	68.75

Group	Body Fat (%)	Fat Mass (kg)	Fat Free Mass (kg)	Actual RV (L)	Pred RV (L)
HE1	13.90	11.2	69.8	2.90	x
HE2	13.00	11.7	78.5	1.71	x
HE3	17.40	13.1	62.2	1.27	x
HE4	13.20	9.6	62.9	1.38	x
HE5	13.00	8.7	58.2	1.18	x
HE6	13.00	8.9	59.8	1.57	x
HE7	7.78	6.8	81.2	1.77	x
HE8	13.50	11.1	71.5	x	1.88
HE9	12.60	11.4	79.0	1.97	x
HE10	8.60	6.7	71.2	1.77	x
HE11	11.70	9.0	68.1	1.41	x
HE12	12.90	9.8	66.0	1.34	x
HE13	12.60	8.6	59.4	1.62	x
HE14	14.00	11.0	68.0	x	1.97
HE15	10.80	8.3	68.5	0.97	x
HE16	10.40	6.9	59.7	1.29	x
ME1	11.30	6.6	51.2	1.01	x
ME2	13.00	9.0	60.0	1.36	x
ME3	11.30	8.6	67.1	1.53	x
ME4	13.50	11.2	71.6	1.03	x
ME5	6.70	4.5	62.0	1.50	x
ME6	13.10	9.5	62.7	x	2.00
ME7	13.10	9.3	61.5	1.10	x
ME8	6.10	3.9	60.4	x	1.90
ME9	22.00	18.5	65.7	1.48	x
ME10	26.70	26.2	71.8	x	1.76
ME11	13.50	9.7	61.6	1.34	x
ME12	16.70	10.7	56.5	1.66	x
ME13	8.30	6.1	67.4	1.59	x
ME14	8.10	5.3	60.5	1.25	x
ME15	10.20	5.7	49.8	1.22	x
ME16	9.00	6.5	65.3	1.67	x
ME17	16.60	13.3	66.9	1.43	x
ME18	8.80	5.7	58.8	1.41	x
ME19	14.00	9.1	55.5	1.71	x
ME20	13.10	11.2	74.9	2.35	x
SC1	24.70	25.0	68.5	1.78	x
SC2	16.90	16.5	80.9	1.26	x
SC3	8.50	9.30	68.8	1.14	x
SC4	24.30	19.2	59.5	1.92	x
SC5	5.80	10.0	65.5	1.00	x
SC6	25.00	25.5	63.5	1.51	x
SC7	12.80	9.05	68.8	1.08	x
SC8	24.80	18.5	55.8	1.53	x
SC9	24.90	20.3	74.5	1.69	x
SC10	5.90	4.50	60.7	x	1.93
SC11	24.90	18.5	55.5	1.53	x
SC12	15.20	15.5	86.3	2.24	x
SC13	24.10	17.5	65.2	1.16	x
SC14	16.40	12.5	62.5	x	1.69
SC15	25.30	21.6	63.3	1.04	x
SC16	16.20	13.5	71.3	2.49	x

Group	VO2 (l/min)	VO2 (ml/kg/min)	VO2 (ml/kgFFM/min)	Max HR (bpm)	VT (L/min)	HR @ VT (bpm)
HE1	5.133	63.2	73.5	188	4.427	168
HE2	4.719	52.3	60.1	188	4.038	164
HE3	4.175	55.3	67.1	184	3.087	161
HE4	4.666	64.2	74.2	188	3.941	188
HE5	3.939	58.8	67.7	197	3.166	159
HE6	3.635	52.9	60.8	196	3.035	180
HE7	5.462	61.9	67.3	185	4.701	162
HE8	5.102	61.6	71.4	199	3.912	187
HE9	5.152	56.9	65.2	182	4.468	169
HE10	4.772	61.3	67.0	175	3.768	150
HE11	5.276	68.3	77.5	179	4.823	154
HE12	4.999	65.8	75.7	197	3.932	176
HE13	4.794	69.2	80.7	181	3.79	166
HE14	5.048	63.5	74.2	176	3.395	157
HE15	4.96	64.5	72.4	193	4.566	172
HE16	4.816	72.2	80.7	200	3.352	173
ME1	3.406	58.6	66.5	197	2.516	162
ME2	4.431	64.1	73.9	186	3.458	166
ME3	4.532	59.7	67.5	196	3.708	171
ME4	5.293	63.9	73.9	187	4.321	172
ME5	4.173	62.6	67.3	193	3.496	175
ME6	4.311	59.1	68.8	180	3.554	171
ME7	4.112	58	66.9	196	3.086	165
ME8	3.51	54.59	58.1	x	2.95	x
ME9	4.822	56.9	73.4	186	3.422	165
ME10	4.281	43.7	59.6	194	3.099	155
ME11	3.55	49.8	57.6	209	2.455	180
ME12	3.32	51.7	58.8	189	2.902	172
ME13	4.658	63.38	69.1	175	3.915	162
ME14	4.601	69.8	76.0	197	3.683	183
ME15	3.447	62	69.2	206	3.199	190
ME16	4.092	55.9	62.7	177	3.538	165
ME17	4.854	60.52	72.6	181	4.21	163
ME18	4.568	70.7	77.7	197	3.548	179
ME19	4.235	65.5	76.3	197	2.958	166
ME20	4.73	54.8	63.2	190	3.618	175
SC1	2.888	39.8	44.5	196	2.348	172
SC2	4.109	42.9	50.8	185	2.917	148
SC3	4.357	50.6	59.6	198	3.536	159
SC4	3.832	47.2	60.2	189	2.767	147
SC5	3.589	40	47.6	188	2.871	150
SC6	3.524	45.7	55.6	204	2.899	188
SC7	3.824	51.4	60	181	2.513	152
SC8	3.545	49.9	63.5	172	2.335	168
SC9	5.095	43.1	55.1	179	2.294	145
SC10	3.749	48	57.6	196	2.913	171
SC11	2.645	35.1	47.5	185	1.848	153
SC12	3.502	48.5	59.5	187	3.196	165
SC13	3.384	40.5	51.7	198	2.856	160
SC14	3.264	43.4	52.8	192	2.568	168
SC15	4.935	63.6	75.6	186	4.218	168
SC16	3.633	41.8	51.0	201	2.586	174

Group	REE (kcal/day)	REE (kcal/kgFFM/day)	Pred REE (kcal/day)	VO2 @ REE (L/min)	RER
HE1	2299	32.9	1756	0.333	0.77
HE2	2424	30.9	1934	0.351	0.77
HE3	1871	30.1	1634	0.286	0.7
HE4	1983	31.5	1713	0.285	0.81
HE5	1860	32.0	1659	0.269	0.78
HE6	1786	29.9	1603	0.274	0.66
HE7	2281	28.1	1930	0.327	0.82
HE8	2077	29.0	1860	0.304	0.73
HE9	2344	29.7	1990	0.343	0.73
HE10	1886	26.5	1794	0.272	0.79
HE11	2240	32.9	1757	0.321	0.82
HE12	2084	31.6	1780	0.299	0.81
HE13	1478	24.9	1709	0.217	0.72
HE14	1977	29.1	1793	0.29	0.71
HE15	2328	34.0	1803	0.339	0.75
HE16	1757	29.4	1643	0.269	0.68
ME1	1644	32.1	1506	0.237	0.79
ME2	1824	30.4	1650	0.279	0.69
ME3	2142	31.9	1768	0.313	0.73
ME4	1975	27.6	1844	0.303	0.66
ME5	1841	29.7	1613	0.283	0.66
ME6	1928	30.7	1674	0.283	0.72
ME7	1907	31.0	1663	0.291	0.7
ME8	1825	30.2	1672	0.267	0.72
ME9	2156	32.8	1799	0.314	0.75
ME10	2148	29.9	2007	0.329	0.68
ME11	1795	29.1	1729	0.262	0.74
ME12	1606	28.4	1544	0.245	0.7
ME13	1963	29.1	1692	0.287	0.73
ME14	2095	34.6	1661	0.321	0.67
ME15	1697	34.1	1460	0.26	0.66
ME16	1956	30.0	1726	0.299	0.68
ME17	2086	31.2	1821	0.318	0.7
ME18	1870	31.8	1640	0.287	0.65
ME19	1743	31.4	1614	0.256	0.71
ME20	2324	31.0	1873	0.34	0.73
SC1	2422	32.4	1914	0.306	0.79
SC2	2241	27.3	1986	0.321	0.76
SC3	2080	27.1	1948	0.305	0.72
SC4	2021	30.8	1764	0.309	0.69
SC5	1511	28.3	1592	0.252	0.66
SC6	1986	31.4	1828	0.291	0.72
SC7	1553	24.5	1765	0.258	0.67
SC8	1848	33.1	1723	0.283	0.69
SC9	2066	27.8	2046	0.317	0.66
SC10	1899	31.3	1751	0.278	0.73
SC11	1786	32.1	1725	0.261	0.73
SC12	2262	28.2	2066	0.329	0.76
SC13	1666	24.5	1898	0.246	0.69
SC14	1912	35.0	1725	0.313	0.76
SC15	1801	28.1	1758	0.276	0.65
SC16	2082	29.2	1864	0.304	0.74

Group	Diet (kcal/day)	Diet (kcal/kgFFM/day)	Ave % Fat	Ave % CHO	Ave % Protein
HE1	3289	47.1	26	59	14
HE2	3509	44.7	23	61	13
HE3	2730	43.9	19	65	16
HE4	2718	43.2	22	61	16
HE5	4098	70.4	22	63	15
HE6	2762	46.2	27	55	16
HE7	4012	49.4	27	54	13
HE8	3292	46.0	29	51	20
HE9	4471	56.6	14	74	12
HE10	2958	41.5	16	67	12
HE11	3616	53.1	30	50	19
HE12	4075	61.7	12	77	11
HE13	3480	58.6	35	49	16
HE14	3465	51.0	32	54	15
HE15	3319	48.5	37	48	15
HE16	3721	62.3	32	52	15
ME1	3551	69.4	22	66	12
ME2	3326	55.4	29	53	17
ME3	3577	53.3	33	53	14
ME4	3654	51.0	21	65	14
ME5	3477	56.1	19	64	15
ME6	2523	40.2	24	59	14
ME7	3048	49.6	20	66	13
ME8	4184	69.3	27	56	16
ME9	2740	41.7	25	44	14
ME10	4199	58.5	20	69	11
ME11	3457	56.1	25	59	14
ME12	2975	52.7	12	71	13
ME13	3194	47.4	28	53	13
ME14	2911	48.1	20	66	13
ME15	3564	71.6	28	50	14
ME16	3768	57.7	31	50	17
ME17	3846	57.5	33	50	17
ME18	3073	52.3	24	60	16
ME19	3368	60.7	36	47	16
ME20	4464	59.6	30	57	13
SC1	2418	36.9	32	40	15
SC2	2905	35.9	33	55	12
SC3	3184	41.5	22	59	18
SC4	2376	45.3	23	62	14
SC5	2762	51.9	28	54	18
SC6	3390	53.6	27	56	14
SC7	2932	46.3	32	49	19
SC8	3144	56.3	29	53	15
SC9	3143	42.3	30	59	11
SC10	3884	51.0	30	42	15
SC11	2088	37.5	32	54	15
SC12	2460	26.9	27	53	19
SC13	3254	49.8	30	57	12
SC14	2963	47.0	29	60	11
SC15	2415	27.0	45	40	15
SC16	2457	34.5	32	48	20

Group	Intens 1 (min)	Intens 2 (min)	Intens 3 (min)	Intens 4 (min)	Intens 5 (min)	Time Exercise (min)
HE1	0	120	298	0	0	418
HE2	0	75	190	175	0	440
HE3	112	0	225	135	0	472
HE4	0	60	62	0	32	154
HE5	0	145	333	70	0	548
HE6	30	0	60	125	0	215
HE7	240	170	145	188	0	743
HE8	0	55	20	60	90	225
HE9	0	40	425	60	0	525
HE10	30	105	0	93	80	308
HE11	331	70	93	0	185	679
HE12	0	0	367	20	0	387
HE13	0	135	90	155	135	515
HE14	68	150	0	60	0	278
HE15	180	45	80	90	0	395
HE16	70	0	424	190	80	764
ME1	0	120	0	128	120	368
ME2	41	12	216	0	82	351
ME3	45	160	207	0	0	412
ME4	0	0	72	135	57	264
ME5	60	0	92	166	0	318
ME6	0	230	319	0	0	549
ME7	0	163	103	0	0	266
ME8	25	0	196	62	0	283
ME9	0	0	408	205	0	613
ME10	0	0	194	343	45	582
ME11	0	20	6	143	0	169
ME12	0	20	237	104	0	361
ME13	0	0	164	122	0	286
ME14	0	114	178	134	30	456
ME15	0	0	240	0	60	300
ME16	390	0	100	100	0	590
ME17	75	78	199	145	0	497
ME18	30	0	180	207	0	417
ME19	0	120	180	225	0	525
ME20	0	55	240	90	0	385
SC1	0	0	0	0	0	0
SC2	0	0	0	60	0	60
SC3	0	0	85	45	10	140
SC4	0	0	40	0	0	40
SC5	50	0	45	0	0	135
SC6	0	0	80	125	0	205
SC7	120	0	135	60	0	315
SC8	40	30	65	0	0	135
SC9	0	0	0	40	0	40
SC10	0	0	0	0	0	0
SC11	0	180	60	30	0	270
SC12	0	150	0	0	0	150
SC13	15	0	0	0	0	15
SC14	0	0	300	0	0	300
SC15	120	120	0	60	0	300
SC16	0	30	0	0	0	30

Group	Ave hrs/wk/year	Reported Training (hrs/week)
HE1	11 to 14	15.5
HE2	11 to 14	18
HE3	11 to 14	9
HE4	11 to 14	21
HE5	11 to 14	14
HE6	11 to 14	8
HE7	11 to 14	12
HE8	11 to 14	14
HE9	11 to 14	12.25
HE10	11 to 14	10.5
HE11	15 +	21
HE12	15 +	21
HE13	15 +	20
HE14	15 +	16
HE15	15 +	16
HE16	15 +	28.5
ME1	5 to 7	5.5
ME2	5 to 7	6.5
ME3	5 to 7	10
ME4	5 to 7	8
ME5	5 to 7	6
ME6	8 to 10	12
ME7	8 to 10	9
ME8	8 to 10	6
ME9	8 to 10	7
ME10	8 to 10	10
ME11	8 to 10	8.5
ME12	8 to 10	8.25
ME13	8 to 10	10
ME14	8 to 10	7
ME15	8 to 10	10
ME16	8 to 10	10
ME17	8 to 10	11.5
ME18	8 to 10	8.5
ME19	8 to 10	8
ME20	8 to 10	10
SC1	2 to 4	0
SC2	0 to 2	0
SC3	0 to 2	1
SC4	0 to 2	1
SC5	0 to 2	1
SC6	2 to 4	5
SC7	0 to 2	2
SC8	2 to 4	4
SC9	0 to 2	1.5
SC10	2 to 4	2
SC11	0 to 2	2
SC12	0 to 2	0
SC13	2 to 4	1
SC14	2 to 4	0
SC15	0 to 2	0
SC16	2 to 4	3

Group	Tritrac -Total kcal	Tritrac - Act kcal	Tritrac - REE kcal
HE1	12519.12	4512.72	8006.40
HE2	12556.84	3542.44	9014.40
HE3	11955.91	4179.91	7776.00
HE4	10821.95	3161.15	7660.80
HE5	10687.04	3199.04	7488.00
HE6	10864.30	3664.30	7200.00
HE7	14105.55	5235.15	8870.40
HE8	11709.37	2997.37	8712.00
HE9	15257.45	5983.85	9273.60
HE10	10752.32	2573.12	8179.20
HE11	11694.53	3860.93	7833.60
HE12	12336.25	4329.85	8006.40
HE13	11317.95	3599.55	7718.40
HE14	11175.41	2996.21	8179.20
HE15	13041.37	4689.37	8352.00
HE16	9741.20	2310.80	7430.40
ME1	10144.53	3405.33	6739.20
ME2	9349.90	1919.50	7430.40
ME3	11100.06	2978.46	8121.60
ME4	11310.50	2958.50	8352.00
ME5	9193.62	2166.42	7027.20
ME6	10495.30	3122.50	7372.80
ME7	9988.13	2557.73	7430.40
ME8	9311.97	1766.37	7545.60
ME9	10955.35	2718.55	8236.80
ME10	11645.39	2285.39	9360.00
ME11	9635.73	1859.73	7776.00
ME12	9489.84	2750.64	6739.20
ME13	10627.57	2909.17	7718.40
ME14	9906.26	2360.66	7545.60
ME15	9966.74	3054.74	6912.00
ME16	11797.86	3906.66	7891.20
ME17	11781.46	3429.46	8352.00
ME18	10546.89	3231.69	7315.20
ME19	10923.06	3665.46	7257.60
ME20	11081.09	2556.29	8524.80
SC1	9793.80	981.00	8812.80
SC2	10714.30	2326.00	9388.80
SC3	11188.36	2317.96	8870.40
SC4	9803.42	1681.82	8121.60
SC5	8699.76	1617.96	7081.80
SC6	10068.43	1658.83	8409.60
SC7	9481.79	1532.79	7949.00
SC8	10386.51	2668.11	7718.40
SC9	10279.80	832.90	9446.40
SC10	9585.00	1636.20	7948.80
SC11	8736.99	1018.59	7718.40
SC12	10923.79	1662.19	9261.60
SC13	10224.74	1584.74	8640.00
SC14	9557.05	1665.85	7891.20
SC15	10148.24	1911.44	8236.80
SC16	n/a	n/a	n/a

Group	Tritrac - ave kcal/day	Tritrac - ave kcal Act/day	Tritrac - ave kcalREE/day
HE1	3130	1128	2002
HE2	3139	886	2254
HE3	2989	1045	1944
HE4	2705	790	1915
HE5	2672	800	1872
HE6	2716	916	1800
HE7	3526	1309	2218
HE8	2927	749	2178
HE9	3814	1496	2318
HE10	2688	643	2045
HE11	2924	965	1958
HE12	3084	1082	2002
HE13	2829	900	1930
HE14	2794	749	2045
HE15	3260	1172	2088
HE16	2435	578	1858
ME1	2536	851	1685
ME2	2337	480	1858
ME3	2775	745	2030
ME4	2828	740	2088
ME5	2298	542	1757
ME6	2624	781	1843
ME7	2497	639	1858
ME8	2328	442	1886
ME9	2739	680	2059
ME10	2911	571	2340
ME11	2409	465	1944
ME12	2372	688	1685
ME13	2657	727	1930
ME14	2477	590	1886
ME15	2492	764	1728
ME16	2949	977	1973
ME17	2945	857	2088
ME18	2637	808	1829
ME19	2731	916	1814
ME20	2770	639	2131
SC1	2488	285	2203
SC2	2929	582	2347
SC3	2976	758	2218
SC4	2451	420	2030
SC5	2175	404	1764
SC6	2517	515	2102
SC7	2370	383	1987
SC8	2597	667	1930
SC9	2570	208	2362
SC10	2396	409	1987
SC11	2182	255	1930
SC12	2741	351	2390
SC13	2556	396	2160
SC14	2389	416	1973
SC15	2537	478	2059
SC16	n/a	n/a	n/a

Group	Tritrac - kcal/kgFFM/day	Tritrac - kcalACT/kgFFM/day	Tritrac -kcalREE/kgFFM/day
HE1	44.8	16.2	28.7
HE2	40.0	11.3	28.7
HE3	48.1	16.8	31.3
HE4	43.0	12.6	30.4
HE5	45.9	13.7	32.2
HE6	45.4	15.3	30.1
HE7	43.4	16.1	27.3
HE8	40.9	10.5	30.5
HE9	48.3	18.9	29.3
HE10	37.8	9.0	28.7
HE11	42.9	14.2	28.8
HE12	46.7	16.4	30.3
HE13	47.6	15.1	32.5
HE14	41.1	11.0	30.1
HE15	47.6	17.1	30.5
HE16	40.8	9.7	31.1
ME1	49.5	16.6	32.9
ME2	39.0	8.0	31.0
ME3	41.4	11.1	30.3
ME4	39.5	10.3	29.2
ME5	37.1	8.7	28.3
ME6	41.8	12.5	29.4
ME7	40.6	10.4	30.2
ME8	38.5	7.3	31.2
ME9	41.7	10.3	31.3
ME10	40.5	8.0	32.6
ME11	39.1	7.5	31.6
ME12	42.0	12.2	29.8
ME13	39.4	10.8	28.6
ME14	40.9	9.8	31.2
ME15	50.0	15.3	34.7
ME16	45.2	15.0	30.2
ME17	44.0	12.8	31.2
ME18	44.8	13.7	31.1
ME19	49.2	16.5	32.7
ME20	37.0	8.5	28.5
SC1	37.6	8.7	28.9
SC2	36.2	7.2	29.0
SC3	36.9	7.9	29.0
SC4	38.6	9.6	29.0
SC5	40.7	7.6	33.2
SC6	39.8	6.6	33.2
SC7	37.4	5.2	32.2
SC8	36.5	12.0	24.6
SC9	34.6	7.8	26.8
SC10	39.5	8.7	32.8
SC11	39.3	7.6	31.6
SC12	34.0	4.2	29.8
SC13	39.1	8.1	31.0
SC14	38.2	8.6	29.6
SC15	39.5	7.5	32.0
SC16	NA	NA	NA

Group	Thinness	Aware	Bulimia	Dissatis	Ineffect	Maturity	Perfect	Distrust	DEBQ-R
HE1	0	2	0	0	0	3	5	0	20
HE2	0	1	0	0	0	5	4	1	24
HE3	1	0	0	0	0	4	0	0	10
HE4	1	0	0	1	0	0	0	2	22
HE5	1	0	2	0	0	1	4	0	13
HE6	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
HE7	0	0	1	3	0	0	1	4	17
HE8	6	7	0	1	1	2	7	5	29
HE9	0	0	0	0	0	6	2	0	17
HE10	3	0	3	0	0	0	3	0	14
HE11	0	1	0	0	2	2	8	0	16
HE12	2	0	0	1	0	3	1	0	30
HE13	0	0	0	0	0	0	1	0	11
HE14	0	0	0	0	0	3	3	0	16
HE15	3	0	5	0	0	2	4	0	22
HE16	0	1	0	3	0	3	8	0	13
ME1	0	1	0	1	1	0	10	2	13
ME2	2	1	0	0	0	0	7	0	24
ME3	0	1	0	1	0	0	10	2	12
ME4	3	0	0	0	3	0	3	0	14
ME5	0	0	0	0	0	1	1	0	18
ME6	0	1	0	3	2	0	1	5	22
ME7	0	0	0	0	2	6	4	11	17
ME8	0	0	2	0	0	4	4	0	13
ME9	7	1	0	11	1	0	4	0	32
ME10	0	0	0	6	0	2	6	2	20
ME11	0	2	0	0	0	6	0	8	14
ME12	0	1	0	0	0	0	4	5	19
ME13	0	0	0	0	0	3	1	0	17
ME14	0	0	0	0	1	1	8	5	16
ME15	0	0	0	7	0	3	2	1	20
ME16	0	0	1	1	0	0	5	1	15
ME17	0	1	0	0	1	0	0	2	17
ME18	0	0	0	1	0	0	1	1	12
ME19	0	0	0	0	1	0	8	0	18
ME20	0	1	0	0	0	1	3	3	13
SC1	2	0	0	2	0	2	3	0	31
SC2	0	0	0	0	0	3	5	1	10
SC3	0	5	0	5	2	1	0	2	3
SC4	0	1	0	3	0	0	3	0	17
SC5	1	0	0	0	1	0	5	5	23
SC6	3	0	0	1	0	1	0	1	23
SC7	0	0	0	0	2	2	3	1	18
SC8	1	0	1	0	0	5	6	5	14
SC9	0	0	0	0	0	0	4	1	24
SC10	0	2	3	0	2	3	3	0	13
SC11	5	0	4	16	12	1	5	8	32
SC12	3	0	0	0	0	2	2	5	25
SC13	0	0	2	0	0	5	6	0	10
SC14	0	0	0	0	2	2	0	0	17
SC15	1	0	0	0	1	0	0	5	24
SC16	1	0	0	3	0	2	4	3	15

Group	Total Test.(ng/dl)	Free Test.(pg/ml)	LH (mIU/ml)	FSH (mIU/ml)	Prolactin (ng/ml)
HE1	301.227	10.294	4.285	9.454	3.487
HE2	726.992	24.239	3.769	2.105	8.106
HE3	319.722	10.963	2.831	2.007	10.752
HE4	435.932	14.192	4.040	2.358	3.210
HE5	294.161	11.321	0.537	3.366	5.949
HE6	553.640	17.456	1.813	4.847	4.652
HE7	533.064	15.733	1.605	1.854	9.983
HE8	557.179	19.016	0.717	1.024	15.964
HE9	450.253	14.331	3.478	2.073	9.012
HE10	268.993	8.515	1.799	3.765	8.998
HE11	305.744	10.021	2.068	2.236	5.747
HE12	298.012	10.432	1.562	0.793	4.933
HE13	386.016	11.225	2.020	2.288	5.440
HE14	435.501	17.878	5.598	5.213	8.699
HE15	338.843	10.016	1.497	2.740	11.082
HE16	376.904	11.634	2.774	5.087	11.794
ME1	350.647	14.080	1.929	3.381	8.940
ME2	349.223	13.215	2.291	8.516	4.993
ME3	614.116	22.591	3.607	3.256	9.522
ME4	329.439	10.694	4.661	3.492	6.173
ME5	267.646	6.085	0.864	2.544	4.433
ME6	672.762	17.119	10.809	13.644	6.441
ME7	417.606	18.437	2.834	1.382	8.395
ME8	304.956	13.181	2.082	1.721	8.252
ME9	330.590	16.676	2.855	2.464	8.166
ME10	312.304	14.330	4.402	1.096	4.283
ME11	396.896	21.739	3.052	0.868	8.402
ME12	275.405	11.604	2.850	6.091	4.482
ME13	510.253	17.176	1.319	1.839	11.260
ME14	658.450	26.686	2.035	4.366	7.070
ME15	348.194	13.895	5.804	2.965	4.151
ME16	675.063	23.136	4.402	2.679	6.032
ME17	707.121	15.789	3.703	8.919	5.311
ME18	387.160	11.401	2.641	2.866	11.289
ME19	529.375	22.847	4.284	3.807	7.759
ME20	256.542	10.613	4.331	4.413	6.647
SC1	640.509	30.142	5.206	4.750	5.627
SC2	369.492	16.575	5.252	4.507	4.043
SC3	453.616	16.512	3.535	4.603	5.425
SC4	416.266	15.562	5.506	5.076	6.886
SC5	410.748	17.347	3.435	2.496	6.168
SC6	642.434	22.165	2.875	3.185	6.532
SC7	478.098	13.363	3.222	4.056	4.745
SC8	302.958	12.729	0.687	1.199	5.601
SC9	619.276	18.368	4.576	4.448	6.869
SC10	428.687	16.850	1.539	4.782	5.745
SC11	616.292	17.021	1.974	5.203	7.815
SC12	390.896	15.220	3.751	1.824	5.822
SC13	458.848	20.347	2.104	2.088	6.844
SC14	495.687	19.638	1.783	3.160	7.104
SC15	285.012	14.361	1.929	3.689	6.553
SC16	640.185	27.185	1.918	2.882	8.833

Group	Cortisol (ug/dl)	Total T3 (ng/dl)	Total T4 (ul/dl)
HE1	13.740	120.018	6.487
HE2	10.817	105.632	6.197
HE3	7.041	97.652	5.757
HE4	9.529	108.047	6.049
HE5	18.351	107.611	6.970
HE6	9.934	100.118	7.101
HE7	8.038	114.278	6.465
HE8	10.038	115.512	7.918
HE9	5.516	109.326	6.159
HE10	11.069	80.650	4.833
HE11	8.632	79.757	6.275
HE12	4.056	93.034	5.746
HE13	10.245	96.614	5.604
HE14	9.880	100.424	6.538
HE15	11.624	96.442	6.743
HE16	5.693	107.060	6.256
ME1	8.266	87.628	5.305
ME2	7.074	113.553	7.570
ME3	9.062	144.172	8.640
ME4	11.366	90.643	5.589
ME5	11.104	92.101	7.203
ME6	8.845	130.516	7.003
ME7	11.874	84.690	5.120
ME8	9.617	110.300	7.773
ME9	8.500	103.395	6.349
ME10	14.231	163.481	8.536
ME11	9.394	103.701	6.030
ME12	8.945	93.936	6.583
ME13	10.555	114.401	8.332
ME14	14.940	110.672	5.758
ME15	9.431	108.364	6.349
ME16	15.803	96.793	5.686
ME17	7.231	114.234	8.197
ME18	17.016	99.654	6.322
ME19	17.365	119.938	6.138
ME20	13.455	121.699	6.631
SC1	17.095	157.453	9.577
SC2	12.437	125.785	6.539
SC3	6.249	92.263	5.997
SC4	6.576	115.102	8.179
SC5	12.210	105.942	5.644
SC6	5.032	137.633	7.302
SC7	12.249	106.128	5.822
SC8	5.579	123.191	6.684
SC9	8.665	123.572	7.856
SC10	3.749	116.666	6.508
SC11	8.076	98.542	6.065
SC12	2.792	109.236	6.307
SC13	3.980	92.435	5.162
SC14	7.283	130.947	7.577
SC15	4.512	107.287	6.682
SC16	18.831	110.712	5.506