REPRODUCTIVE AND ENDOCRINE PARAMETERS OF FAT

VERSUS MODERATELY CONDITIONED MARES FOLLOWING

PARTURITION

A Dissertation

by

CLAY ALAN CAVINDER

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

August 2006

Major Subject: Animal Science

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Approved by:

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ABSTRACT

Reproductive and Endocrine Parameters of Fat versus Moderately

Conditioned Mares Following Parturition. (August 2006)

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An increase in time to ovulation following parturition could result in economic loss if the mare cannot successfully conceive within a short time after foaling. To evaluate if a difference exists in reproductive efficiency of fat- (body condition score of 7 to 8) versus moderately-conditioned (body condition score of 5 to 6), 24 mares were allotted to and maintained in their respective group from late gestation until pregnancy was confirmed following breeding on the second post-partum estrus. Days to ovulation, interovulatory intervals, conception rates, and endocrine profiles were analyzed. Serum concentrations of thyroxine (T_4), insulin-like growth factor-1 (IGF-1) and leptin were assayed in order to characterize normal circulating blood concentrations. There were no differences (P>0.05) in mean interval from parturition to first ovulation (14.41 $\pm$ 1.07 and 16.18 $\pm$ 1.06 d), first to second postpartum ovulation (22.91 $\pm$ 1.07 and 24.33 $\pm$ 0.93 d), or in conception rates (91.67% and 83.33%) between the 2 groups. However, mares in moderate conditioning did lose a greater percentage of body fat upon foaling as compared to fleshier mares (0.82% versus 0.35%). Leptin concentrations were not different between the groups (P>0.05). Nevertheless, serum concentrations of T_4 were

higher (P<0.01) and IGF-1 concentrations lower (P<0.01) in moderate- as compared to fat-conditioned mares during times of ovulation and the interovulatory period. Results indicate that mares maintained in a fleshy body condition are not prone to reproductive dysfunction or lowered levels of fertility. The significance of the current results is important as it reassures the breeder that mares in a fatter body condition score (BCS of 7-8) should not demonstrate sub-fertility related to level of body fat. Additionally, results indicate that mares may need to be kept in a BCS of 6 in order to avoid losing enough weight upon parturition and early lactation to bring the BCS below 5. It suggests that varying amounts of circulating T_4 and IGF-1 do not affect reproductive capabilities of mares in a BCS of greater than 5 following parturition.

DEDICATION

To my wife

ACKNOWLEDGEMENTS

I have always been proud of the fact that I am a person who is motivated and not scared of putting forth hard work in order to be better than average at what I decide to pursue. The pressure that I put on myself may even be one of my weaknesses. However, through the process of attaining my doctorate I have come to realize that my goals and aspirations are not only reached by my own drive. My goals are more importantly assisted by the people who love and care about me. I know I couldn't have gotten this far without the love, support and encouragement of a few people. First of all, I have had the honor of working with a great person, teacher and mentor in Dr. Martha Vogelsang. She has encouraged me and given me the confidence I need to be a great teacher and scientist. Dr. Pete Gibbs is someone I look up to also. He has shown me something outside the realm of science. He has shown me something very simple; how to be a person in a leadership role with integrity and character while at the same time being a "straight shooter." Furthermore, Dr. David Forrest and Dr. David Schmitz have been wonderful in lending their support and giving great advice through the writing process. Thank you all for everything!

I have many graduate and undergraduate students to also thank. This project was fairly large and required the assistance of 21 student assistants and many extra hours from friends. Thanks to Tommy Chancellor, Betsy Wagner, Elena Eller, and Chris Mortensen for their helping hands.

In the past 3 years I have learned a lot about being a good teacher, a dedicated student, and a motivated coach but nothing has been as important as becoming a better

husband. Ginger is so deserving of everything she has and more. She has ALWAYS been there for me and given me the love I need to be successful in my work. Many nights I have been discouraged with school and she has looked past my grouchy attitude to lend me a hand of support. Even though the last few years have been hard, I will always cherish them for how they have brought us closer and shown us that we can work through anything. You are my best friend.

I have saved the most important for last: Jesus Christ. I cannot imagine where my life would be without knowing him and depending on his grace and mercy each and every day. Life is the hardest when I am not as close to him as I should be. When I call on him, he is always there giving me the strength I need. I know without a doubt that I couldn't have completed this task without him opening the door of opportunity first and then helping me walk the path he has chosen for me. Also, I can't thank him enough for giving me the opportunity to work with his creation the horse. I have a job that I enjoy and look forward to doing because of the love of horses he has given me. Lord, I thank you for giving me that desire and pleasure in this life. The word says in Psalm 37:4, "Delight yourself in the Lord and he will give you the desires of your heart." This is just one of many blessings you have given, thank you.

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CHAPTER I

INTRODUCTION

Mares have a relatively long gestation (approximately 340 days) that requires

rebreeding soon after parturition to insure the birth of a foal the following year. Unlike

other animals, the mare has an early postpartum estrus that begins around d 7 to 10 after

foaling, and is referred to as the foal heat (Fitzgerald et al., 1985). In order to achieve an

early pregnancy following parturition, breeding the mare on the foal heat or having her

quickly return to estrus following foal heat is desired.

It is apparent that an adequate amount of body fat is necessary for females to

achieve normal reproductive processes. Therefore, a body condition following

parturition that is most beneficial for achieving a quick pregnancy in the mare is

necessary. The relationships among nutritional status, body condition, and reproductive

efficiency have been studied in the horse (Henneke et al., 1984; Hines et al., 1985), cow

(Randel, 1990), and the ewe and sow (Dunn and Kaltenbach, 1980). The

aforementioned reports indicate that extreme over- or under- nutrition can cause delays

in reproductive function. Obesity has been related to reproductive problems in humans

and animals. An abnormally long estrous cycle is one example of an impairment

associated with obesity in humans (Hartz et al., 1979), and has been speculated to impair

reproductive function in the mare (Fitzgerald et al., 2003). However, Kubiak et al.

(1989) concluded that a high degree of body fat (Body Condition Score = 9) produced by

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This dissertation follows the style and format of the Journal of Animal Science.

overfeeding during gestation did not adversely affect postpartum reproductive performance in mares. Studies have also concluded that females who are too thin as a result of inadequate nutrition may suffer from reproductive inefficiencies such as: longer interovulatory intervals, decreased pregnancy rates, and decreased ovarian activity (Dunn et al., 1969; Rutter and Randel, 1983; Richards et al., 1986). Research comparing mares in obese body condition to those in a moderate body condition is limited with the majority of research comparing mares of fat condition to those of thin condition. Since it is accepted that a low body condition impairs reproductive function, a comparison of moderate- to fat- body condition would give horse breeders a more ideal nutritional range in which to manage the broodmare. A body condition that allows the owner to minimize feed costs, while maximizing reproductive potential is the most ideal and beneficial to the equine industry.

Control of the estrous cycle by the level of fatness the mare possesses is mediated by the endocrine system. Leptin is a hormone that is secreted from adipocytes and may serve as a signal from fat cells to the brain which reflects the adequacy of fat stores for reproduction (Bray, 1996). Also, the thyroid gland is suspected of affecting the estrous cycle through secretion of thyroxin (T_4). Thyroidectomized ewes have an extended breeding season and termination of the breeding season by decreases in serum LH can be seen in females treated with exogenous T_4 (Dahl et al., 1995). Furthermore, insulin-like growth factor-1 (IGF-1) is regulated by growth hormone (GH) and plays a role in maintenance of body weight (Tannenbaum et al., 1983). Insulin-like growth factor-1 is higher in females maintaining a higher body condition and has a positive

correlation with LH pulsatility (Beam and Butler, 1999). IGF-1 is also thought to play a key role in the ovaries' sensitivity to follicle stimulating hormone (FSH). These factors likely contribute to the efficiency with which the mare cycles and conceives after foaling. Comparison of the various serum concentrations of these hormones in the mare will provide more understanding of the reproductive control they exert over the mare's reproductive system.

The aim of this study was to determine whether there is a difference in reproductive efficiency in mares that are in a fat- versus moderate- body condition. Comparisons of reproductive efficiency were ascertained by analyses of the intervals from parturition to foal heat ovulation and the second postpartum ovulation, along with calculation of the interovulatory period. Furthermore, pregnancy rates of mares in these 2 groups were compared. Therefore, the objectives of this study were to:

1. compare rebreeding efficiency by determining the length of time from foal heat ovulation to the second postpartum ovulation (or interovulatory period) and to calculate pregnancy rates between mares in fat- (BCS of 7 to 8) versus moderate- (BCS of 5 to 6) condition, and

2. analyze the blood serum concentrations of LH, progesterone, leptin, T_4, and IGF-1 between mares in fat- versus moderate- condition.

The results of this study will provide information necessary for making recommendations to the horse breeding industry on feeding programs. This may

enhance the reproductive efficiency of broodmares, and ultimately, their economic

potential as it relates to foal production.

CHAPTER II

REVIEW OF LITERATURE

Body Condition

Establishment of a body condition scoring system, which applies a numerical score to the amount of body fat an animal possesses, has been achieved in cattle (Whitman, 1975) and horses (Henneke et al., 1983). The body condition score (BCS) is a numerical score with 1 indicating extremely emaciated and 9 indicating extremely fat. It is based on the visual and palpable appraisal of the amount of fat covering on the body at certain locations such as the ribs, tailhead, withers, neck, behind the shoulder, and along the back. Body condition is related to reproductive function in numerous species including humans. Studies in women have found that a certain threshold level of fat storage is needed for menstruation to occur (Frisch and McArthur, 1974). Women with low body fat, such as ballet dancers and long distance runners, have fat levels below this threshold, and subsequently cease menstruating. Furthermore, menstrual irregularities linked to excessive amounts of fat have been corrected with weight loss (Bray, 1996). Additional problems associated with obesity in women are prolonged labor (Calandra et al., 1981), increased incidence of induced labor and the tendency for emergency cesarean sections during delivery (Ekbald and Grenman, 1992). Undernutrition and overnutrition can reduce placental and fetal growth and can even result in intrauterine growth retardation (Wu et al., 2004).

There are numerous studies comparing body condition and the estrous cycle of the mare. Fitzgerald et al. (2003) stated that mares of a BCS of 7 or higher had

lengthened luteal phases and enhanced length of interovulatory periods as compared to lean mares in a BCS of 4. Contrarily, Kubiak et al. (1989) reported that the intervals from foaling to first cycle ovulation, second cycle ovulation and first to second cycle ovulation, were similar between mares in a BCS of 5.5 to 7 versus mares in an overly obese body condition of 9 with all mares achieving and maintaining pregnancy. They concluded a high degree of body fat produced by overfeeding during gestation did not adversely affect postpartum reproductive performance in the multiparous mare. Studies have confirmed the consequences of mares being in a low versus high body condition are longer mean gestation lengths (Hines et al., 1987), longer intervals from parturition to first ovulation, lower pregnancy rates to 30 days post-ovulation, and reduced pregnancy maintenance to 90 days (Henneke et al., 1984). To complement the above results, Henneke et al. (1984) found that mares entering the breeding season or foaling at a BCS of 5 or higher had enhanced breeding efficiency. In comparison, cows in a BCS of less than or equal to 4 as compared to greater than or equal to 5 have lowered pregnancy rates by as much as 30% (Rae et al., 1993) and if not lactating, can undergo anestrus (Richards et al., 1989). DeRouen et al. (1994) also found that cows calving in a BCS of 6 to 7 had higher pregnancy rates compared to those with BCS of 4 to 5 with the interval from parturition to pregnancy for cows calving at a low BCS being 10 to 18 days longer than for those of a higher BCS.

Body condition score at parturition has been implicated as the single most important factor affecting postpartum interval to estrus and pregnancy rate in cows (Richards et al., 1986). Cows calving in a BCS of 4 or lower and that lost weight have

lower pregnancy rates by 20, 40, and 60 days post parturition as compared to cows with a BCS of 5 or higher (Richards et al., 1986). Dunn and Kaltenbach (1980) found that body condition at parturition had a significant effect on the length of postpartum interval. Cows in good body condition at calving exhibit estrus by day 60 regardless of whether they gained or lost weight before or following parturition. Whitman (1975) found that energy restriction in late prepartum cows lowers body condition at calving, thus resulting in an increased interval from parturition to the first postpartum estrus. Feeding high protein diets to dairy cattle seems to shorten the postpartum interval. The postpartum interval is defined as the time from which the female gives birth until the first postpartum estrus which is accompanied by ovulation. Research shows that cows in moderate to thin body condition exhibit estrus by 60 days after parturition if the cows gain weight prior to calving versus those that lost weight prior to parturition (Dunn and Kaltenbach, 1980).

Nutritional status has been implicated in hypothalamic, pituitary and ovarian function (Goater et al., 1981) and maintenance of body condition following parturition has also been shown to increase pituitary function thus increasing reproductive potential. Researchers analyzed body condition from parturition to 20 days postpartum and found cows maintaining body condition had shorter postpartum intervals, higher luteinizing hormone (LH) levels, and higher peaks of LH when exogenous doses of gonadotropin releasing hormone (GnRH) were administered. Additionally, 88% percent of females that were able to maintain body condition after parturition achieved estrus within 42 days postpartum compared to only 36% of cows that were unable to maintain body

condition after calving (Rutter and Randel, 1983; Houghton et al., 1990). Richards et al. (1989) investigated nutritional anestrus in beef cows. Cows of moderate to good body condition were separated into 2 groups and randomly assigned to either a maintenance diet or a restricted diet. Cows on maintenance diets were fed to maintain body condition, and those on the restricted diets were fed to lose 1% of their body weight until luteal activity stopped. Once luteal function ceased, the diets were increased to add body weight to the cows until ovarian activity resumed. They concluded anestrus occurs when nonlactating cows lose weight and achieve a BCS of approximately 3.5. As a result of anestrus, the LH pulse frequency is decreased. When the nutrient intake of the cows was increased after a period of restriction, estrous activity and normal pregnancy rates were re-established. This suggests that greater body fat reserves are required to reinitiate estrous cycles.

Experiments have also been conducted analyzing varying levels of energy intake on reproductive efficiency. Henneke et al. (1981) examined postpartum reproductive performance in mares fed different levels of energy prior to parturition. The parameters analyzed were length of gestation and interval from parturition to first and second ovulation. The diets did not influence gestational lengths, or the interval from parturition to first ovulation. However, the interval from foaling to second ovulation was lengthened in mares foaling in a thin body condition versus mares foaling in a fat body condition. Further findings were that mares in a BCS of less than 5 at foaling had lower pregnancy rates and maintenance of pregnancy to 90 days as compared to mares who foaled at a BCS of 5 or higher. In addition, Houghton et al. (1990) examined cows

that were in moderate body condition 190 days prepartum and then fed them one of two diets; a low or high energy diet. Cows fed low energy were in a thin body condition at calving and had longer postpartum intervals, but higher first service conception rates when compared to moderate and fleshy cows. Thin cows exhibited an extended postpartum anestrous period between 28 and 58 days longer than that of moderate to fleshy cows. Wiltbank et al. (1962) fed pregnant cows varying levels of energy before and after parturition. Conclusions were that cows fed low energy before parturition and high energy diets after calving had significantly longer intervals from calving to first estrus versus cows fed high energy levels before parturition. The effects of the low energy levels before parturition were not overcome by feeding high energy levels after parturition, further substantiating that body condition is an important factor affecting the interval from calving to first estrus.

Ovarian activity can be affected by body condition. Gentry et al. (2002a) evaluated the ovarian activity of fat- and thin- body conditioned mares. These researchers concluded that mares with high body condition (BCS = 6.5 to 8.0) had larger ovaries and more corpora lutea (CL) than mares with low BCS (BCS = 3.0 to 3.5). Mares in high body condition also had larger follicles, higher plasma leptin concentrations, and produced higher daily LH concentrations. Mares with high body condition continued to have abundant ovarian follicular activity. Furthermore, low body condition in mares was associated with a consistent seasonal anovulatory state.

The amount of digestible energy (DE) fed to pregnant mares in late gestation is suggested by the National Research Council (NRC) to be between 12% and 19% over

that required for maintenance. Goater et al. (1981) studied the amount of DE fed to mares 90 days before foaling. One group of mares was fed 100% of the NRC requirements of DE and the other group of mares was fed 120% of the NRC requirements of DE for mares in late gestation. No differences were detected between treatment groups in length of gestation and birth weight of the foals, but the average daily gain of mares fed 120% of the NRC requirements for DE was higher. Also, mares in the higher DE treatment group were better able to maintain postpartum body weight. However, no effect of dietary treatment was observed on progesterone and LH levels in the blood.

Further evidence of a need for an adequate body condition in mares at the time of foaling was observed by Powell et al. (1989) as they evaluated body fat store changes in broodmares during the second and third trimesters of gestation. An ultrasonic scan of the fat over the rump (Kane et al., 1987) was used to predict total body fat percentage. Body weight remained relatively constant in late gestation, but BCS, rump fat thickness and estimated percentage body fat declined. The mares were mobilizing body fat to keep up with the demands of the growing fetus. Also, researchers found that non-lactating mares were more reproductively efficient than lactating mares of the same body condition; as lactating mares put energy reserves towards milk production. Consequently, reproductive performance was enhanced when lactation ceased. A body condition score of 5 appears to be only marginally acceptable and does not meet the stored body fat requirement of the lactating mare on pasture (Gibbs and Davison, 1992).

The Estrous Cycle: LH and Progesterone

Progesterone promotes the establishment of a favorable uterine environment for survival of the developing embryo and is thus critical for successful pregnancy. Progesterone is primarily secreted from the corpus luteum (CL) of the mare's ovary (van Rensburg and van Niekerk, 1968). Studies have shown that, unique to the mare, LH concentrations remain elevated following ovulation. These elevations in LH after ovulation may be important for the development of the CL (Noden et al., 1975). When antibodies for LH were given to mares, CL weights were less than that of control mares (Pineda et al., 1972). This indicates that endogenous pituitary factors which were inhibited by the antiserum, presumably LH, were necessary for development of the CL. However, during times of high concentrations of progesterone, LH concentrations are depressed and the mare is either pregnant or in a period of diestrus (Garcia and Ginther, 1978).

Progesterone is usually below the detectable concentration for radioimmunoassay (RIA) when the mare is in estrus (Plotka et al., 1975). Progesterone secretion from the CL of the mare occurs earlier than in other farm animals which results in systemic levels rising as early as 24 to 36 hours after ovulation to approximately 4 to 22 ng/mL by 5 to 7 days after ovulation. In the nonpregnant mare, the CL continues to produce progesterone until approximately 13 or 14 days after ovulation (Nett et al., 1979). At this time, the uterus secretes prostaglandin to lyse the CL, progesterone declines and the mare comes into estrus. Evans (1991) conducted a study to outline the pattern of progesterone secretion during various stages of the equine estrous cycle and to determine

the correlation of progesterone and LH concentrations. Blood samples were drawn at 3

min intervals for 2 h and at 15 min intervals for an additional 6 h from mares in early (d

3), mid (d 7), and late (d 15) diestrus. Ultradian rhythms of progesterone were detected

in the 3 min and 15 min samples in all mares except those that had experienced

luteolysis prior to d 15 postovulation. The average number of secretory episodes was

3.6 and 3.5 for the 8 h period. The average inter-peak interval and the average peak

duration of progesterone was less for the d 7 group when compared with the d 3 and d 15

groups. The d 15 groups had larger maximum heights and average peak heights of LH

than the other 2 groups. This study concluded that the pattern of progesterone secretion

changes during different stages of diestrus.

Progesterone can have both an inductive and suppressive effect on the secretion

of GnRH. The stimulatory action of acute increases in progesterone may contribute to

the LH surge in humans (Nippoldt et al., 1989). The most common effect of

progesterone on the generation of LH is via a negative-feedback mechanism. During the

luteal phase of the menstrual cycle, plasma progesterone and estrogen levels are high

and pulse frequency is slow. LH pulse frequency rises during the follicular phase when

plasma estrogen and progesterone are lower. This inverse relationship between LH

pulse frequency and circulating levels of progesterone has also been reported in the cow

(Rahe et al., 1980), sheep (Karsch, 1987), and horse (McKinnon and Voss, 1993). For

example, Humphrey et al. (1983) studied hormonal patterns during postpartum anestrus

in beef cows. The authors concluded that the endocrine events associated with the

occurrence of the first postpartum estrus appear to be increased episodes of LH secretion

followed by short lived (2 to 3 d) secretions of progesterone. The secretion of LH and progesterone was then followed by a pre-estrous secretion of estradiol-17β and an ovulatory surge of LH.

Undernutrition results in smaller amounts of fat accumulation and has been shown to directly affect the estrous cycle. A negative energy balance can impair the development of the ovarian follicle and CL and result in less progesterone in serum (Spicer et al., 1990). Folman et al. (1973) examined the relationship between plasma progesterone levels and conception in post-partum dairy cows maintained on a diet of either low or high nutrition. Cows maintained on a high level of nutrition required fewer inseminations per conception, conceived earlier and had a high plasma progesterone level 23 days earlier than cows maintained on a low level of nutrition. The authors suggest that plasma progesterone concentration during the estrous cycle before insemination is closely related to the occurrence of conception. Rhodes et al. (1996) monitored endocrine and ovarian changes directly before the onset of nutritionally induced anestrus in heifers. Anovulatory and initial estrous cycles of feed-restricted heifers were compared with heifers fed ad libitum. These researchers found that feed-restricted heifers had lower concentrations of LH, and the diameter of dominant follicles was smaller, but concentrations of FSH were higher versus heifers fed ad libitum. Reduced dietary intake was associated with insufficient circulating LH to stimulate complete growth of the ovulatory follicle which resulted in ovulation failure. A study analyzing the effects of undernutrition on postpartum rebreeding of cows found that restriction of dietary energy late in the prepartum period or early in the postpartum

period reduced the number of cows returning to estrus within a defined breeding season (Randel, 1990). The effects of dietary energy restriction were attributed to the failure to develop preovulatory ovarian follicles. Similarly, feed restriction in ewes during the luteal phase of the estrous cycle can diminish the magnitude of the preovulatory LH surge release (Kiyma et al., 2004). Hines et al. (1987) evaluated reproductive efficiency in mares in a BCS of 4.5 or less (thin) versus mares in a BCS of 6 or greater (control). Thin mares had irregular periods from parturition to ovulation, while the control mares demonstrated more uniformity in the estrous cycle following parturition. Thin mares also did not appear to respond to natural increases in photoperiod, exhibiting no correlation between time of year and interval from foaling to either the first or second estrous cycle. Thompson et al. (1986) found that serum LH concentration was somewhat dependent upon photoperiod and progesterone level. In the summer, concentrations of LH in serum were higher in mares with low serum progesterone concentrations as compared to mares with high progesterone concentrations. Furthermore, Gentry et al. (2002b) found that even in the winter month of January mares in high body condition exhibited higher progesterone levels, a greater number of corpora lutea, and more large- and medium-size follicles as compared to mares in low body condition.

Leptin

Leptin is a potent satiety hormone synthesized and secreted by adipocytes and is highly correlated with body weight and adiposity (Barash et al., 1996; Cammisotto and

Bukowiecki, 2002; Garcia et al., 2002). The lack of leptin production by adipocytes in humans causes an infrequent form of hunger and obesity (Montague et al., 1997). Studies have shown that leptin concentrations fluctuate with body weight changes as a 10% reduction in body weight causes a reduction of 53% in serum leptin concentration (Considine et al., 1996), and a 10% increase in body weight correlates to a 300% increase in serum leptin concentration in humans (Kolaczynski et al., 1996). Leptin is even thought to be involved in the development of puberty in heifers (Garcia et al., 2002), rodents and humans (Kiess et al., 1999). Prior research gives evidence that leptin acts as a signal triggering puberty as fat accumulation enhances maturation of the reproductive tract (Chehab et al., 1997).

Many studies on leptin and its relationship to metabolism, physiology, and obesity have been conducted with the aid of *ob/ob* mice. These mice have a recessive genetic obesity that is attributed to an absence of leptin secretion from the adipocytes and results in sterile adult mice with over 50% body fat (Yu et al., 1997; Houseknecht et al., 1998). Additional strains of mice have been used that have a genetic resistance to leptin and are referred to as *db/db* mice. In *ob/ob* mice leptin reduces food intake, decreases insulin concentration and lowers blood glucose concentration when administered exogenously, but not in the *db/db* mouse because of their resistance to leptin (Bray, 1996). Injection of recombinant mouse leptin into *ob/ob* mice has been found to lower food intake by 56% and body weight by 4.1% (Schwartz et al., 1996).

Observations have shown that leptin plays a role as an inhibitor of insulin secretion (Kieffer et al., 1997) and research has revealed that insulin approximately

doubles the basal rates of leptin release from adipocytes (Cammisotto and Bukowiecki, 2002). Emilsson et al. (1997) found that leptin acts through the functional leptin receptor to inhibit insulin secretion. They conclude that overproduction of leptin, especially in abdominal fat, may modify insulin secretion directly and could be involved in the development of the diabetic syndrome. The failure of leptin to inhibit insulin secretion of *ob/ob* and *db/db* mice may explain the development of hyperinsulinemia, and insulin resistance seen in these strains of mice. Muller et al. (1997) found that leptin impairs a broad spectrum of insulin actions in rat adipocytes. Rat adipocytes are highly sensitive to insulin, and leptin has several important metabolic effects on insulin including stimulation of glucose transport, glycogen synthase, lipogenesis, and protein synthesis as well as inhibition of iso-proretenol-induced lipolysis. Leptin treatments given to *ob/ob* mice, or diet-induced rats, have been shown to dramatically improve glucose metabolism and insulin sensitivity. In lean rats, leptin injections stimulate whole-body glucose turnover (Muoio and Dohm, 2002). Insulin is involved in increasing leptin secretion in cats, as it is in other species. Appleton et al. (2002) studied the effects of leptin on insulin secretion in cats and found that in both lean and overweight cats, the higher the leptin concentrations, the more insulin resistant the cat, independent of the degree of adiposity. They concluded that leptin overproduction may contribute to the diminished insulin sensitivity seen in overweight cats and humans or alternatively, the compensatory hyperinsulinaemia found with insulin resistance could stimulate leptin production.

Leptin may serve as the signal from fat to the brain regarding the adequacy of fat stores for reproduction (Bray, 1996) and is involved in the hypothalamic-pituitary axis under defined nutritional conditions in cattle (Williams et al., 2002). Sir-Petermann et al. (1999a) proposed that fluctuations in insulin and glucose concentrations drive the ultradian leptin secretion, which in turn is coupled to the LH pulsatile secretion. Barash et al. (1996) postulated that leptin may serve as a metabolic signal to the reproductive system, informing it that adequate fat stores are available to meet the caloric demands of reproduction. Metabolic stresses such as severe exercise and food restriction would signal the reproductive system via low circulating concentrations of leptin, indicating that the resources needed for reproduction were unavailable. Additionally, Yu et al. (1997) hypothesized that when fat stores reach a critical point, there is increased release of leptin from adipocytes into the bloodstream. Leptin then acts on hypothalamic cells to stimulate release of luteinizing hormone-releasing horomone (LHRH), thereby triggering gonadotropin release. In *ob/ob* mice, improvements to reproductive capability and endocrine status, reduced food intake and weight loss are all evident when exogenous doses of leptin are administered (Houseknecht et al., 1998). In a study with hamsters, Schneider et al. (1998) induced anestrus by fasting and then injected the animals with intraperitoneal doses of leptin. Results indicated that 77% of fasted, leptin-treated hamsters exhibited estrous cycles in length of four days compared to 0% of the fasted, vehicle-treated animals. Leptin treatment significantly decreased estrous cycle length. Fasting-induced anestrus was reversed and normal sexual and social behavior and ovulation rates were restored with exogenous leptin. Similarly, Nagatani et al.

(2000) examined the control of pulsatile LH by fasting sheep and found that endogenous levels of leptin decreased during the fast. Vehicle-treated sheep exhibited a decline in LH pulsatile secretions during fasting and exogenous leptin prevented a decline in LH pulse frequency. However, Nagatani et al. (1999) demonstrated that leptin administration to fed rats did not affect pulsatility of LH. This study concluded that the effect of leptin on pulsatile LH secretion is only apparent in hypogonadotropic, fasted animals with low leptin concentrations. Williams et al. (2002) found that short-term fasting of growing peripubertal heifers caused a marked decrease in leptin gene expression and circulating leptin, concomitant with a reduction in LH pulse frequency, and serum concentrations of insulin and IGF-1. Additionally, significant and rapid decreases in leptin concentrations were found in feed-restricted mice as compared to fed mice and this rapid decrease was accompanied by delayed estrus in females (Ahima et al., 1996). They also concluded that LH was undetectable in fasted mice and that leptin restored levels to nearly 40% of those in fed controls, therefore exerting some control on the hypothalamic-pituitary system. Furthermore, changes in adrenal and thyroid axes induced by starvation were influenced by leptin. Research evaluated the effect of a single day of feed restriction on gonadotropin and leptin levels in the mare. In feed-restricted mares, serum leptin concentration decreased, but serum leptin concentration was not altered in the control-fed mares. Prolactin, FSH, and LH serum concentrations were not significantly altered by feed restriction. The absence of suppression of these reproductive hormones may reflect the maintenance of adequate levels of metabolizable

fuels, rather than a failure of leptin to signal nutritional status to the reproductive axis of the mare (McManus and Fitzgerald, 2000).

A pattern of leptin and LH release has been demonstrated (Sir-Petermann et al., 1999b). The nocturnal rise in leptin may determine the nocturnal LH profile in mid-to-late follicular phase that precedes ovulation in women. Studies in the human and rat have demonstrated that leptin secretion follows a diurnal rhythm, rising nocturnally to peak around midnight and the early morning, before declining gradually to reach a late morning or early afternoon nadir (Saad et al., 1998). Licinio et al. (1998) sampled plasma from the blood of healthy women during their menstrual cycles. Leptin concentrations were elevated at night and LH pulsatility significantly changed from low amplitude, high frequency to high amplitude, low frequency becoming synchronous with leptin. These researchers postulated that leptin is not only a trophic factor for the reproductive system, but that the pulse patterns as well as absolute concentrations of leptin, may organize the optimal minute-to-minute functioning of the hypothalamic-pituitary-ovarian (HPO) axis.

Serum concentrations of leptin increase as fat mass increases in horses (Buff et al., 2002). Gentry et al. (2002b) determined the effects of high versus low body condition scores on reproductive characteristics, hormonal secretion, and leptin concentrations resulting from feed restriction in mares. These researchers found that mares with high body condition exhibited greater plasma leptin concentrations and were more responsive to exogenous GnRH than mares in low body condition. Heidler et al.

(2002) found that long term energy losses, such as during lactation, and decreased glucose levels are associated with a clear decrease in leptin concentrations.

Leptin interacts with other hormones in many different species of animals. Ahima et al. (1996) found serum thryoxine decreases with fasting and that leptin administration decreases the fall of T_4. This study demonstrates that falling leptin concentration is a signal initiating the neuroendocrine response to starvation, including limiting procreation, and decreasing thyroid hormones. Furthermore, in-vitro systems provide evidence that growth hormone (GH) can modulate leptin expression, presumably by alterations in tissue response to hormones such as insulin in cattle (Houseknecht et al., 2000). Obese mares have been found to have higher insulin and leptin concentrations and lower thyroxine concentrations as compared to lean mares (Fitzgerald et al., 2003). Cartmill et al. (2003) evaluated hormonal interactions in fat horses (BCS of 7.5 or higher) and found that horses with high leptin concentrations exhibited high concentrations of insulin and triiodothyronine (T_3) but low levels of GH as compared to horses having low levels of leptin in the blood. Horses with high leptin had a greater insulin response to intravenous (i.v.) glucose infusion than horses with low leptin. Similarly, Yu et al. (1997) injected leptin into female rats that had been ovariectomized. Leptin induced a significant increase in plasma LH, which peaked 10 to 50 min after injection of leptin.   Amstalden et al. (2002) administered ovine leptin to cows and found that it significantly stimulated pancreatic insulin and pituitary LH secretion in mature, fasted cows. Thus, leptin may play an important role as a signal linking nutritional status to the central reproductive axis in cattle.

Photoperiod may also be important in leptin secretion. Fitzgerald and McManus (2000) examined the photoperiod versus metabolic signal in the mare and its relationship to seasonal anestrus. A determination was made in both young and mature mares that body weight, percentage body fat and circulating concentration of leptin are higher during the summer as compared to the winter. The mare becomes anestrus in the short days of winter due to an increase in melatonin. This melatonin secretion is modified by energy availability, which may be signaled to the hypothalamic-pituitary axis by a change in the circulating levels of leptin (Fitzgerald and McManus, 2000). They suggest that the occurrence of seasonal anestrus in the mare is determined, to some extent, by metabolic signals.

Thyroxine

Thyroxine has been suggested to play a role in seasonal reproduction. The transition to anestrus is actively driven by the seasonal increase in thyroid hormone secretion in ewes (Karsch et al., 1995). Research has shown that the breeding season of ewes can be extended by lowering thyroxine secretion (Follett and Potts, 1990), or that it can be terminated by exogenously administering large doses of thyroxine (O'Callaghan et al., 1993). Thyroidectomy of mature ewes was found to block the transition from the breeding season to anestrus and to cause ewes to exhibit estrous cycles throughout the year (Moenter et al., 1991; Webster et al., 1991; Karsch et al., 1995). Studies have also shown that thyroid hormones act permissively, only their presence needed to end the breeding season (Shi and Barrell, 1994; Dahl et al., 1995; Karsch et al., 1995). Dahl et

al. (1995) thyroidectomized (THX) ewes late in the anestrous season. Thyroxine was then exogenously replaced or not given to ewes. In all groups, LH increased in mid-September, signifying that manipulation of thyroid status did not influence onset of the neuroendocrine transition to estrus. Additionally, LH remained elevated throughout the study in THX ewes not receiving exogenous thyroxine. Similarly, Moenter et al. (1991) monitored LH secretion in THX and thyroid intact ewes. Luetinizing hormone concentration rose in response to the natural breeding season in both sets of ewes. However, a marked difference in the pulse frequency of LH occurred at the end of the breeding season. Luetinizing hormone concentration declined to basal levels in the thyroid intact ewes and remained elevated in the THX ewes. These observations lead to the conclusion that the thyroid is necessary for endogenous suppression of neuroendocrine mechanisms that generate LH pulses, a suppression crucial for the transition to anestrus.

The level of secretion of thyroxine by the thyroid is also affected by estrous cycle status and meal frequency. Johnson (1986) stated a seasonal change in the concentration of T_4 exists in the mare, but found no change in T_4 during the estrous cycle. Kelley et al. (1974) also concluded no change in thyroxine concentrations in the estrous cycle of the mare except that thyroxine levels declined after ovulation. Amino et al. (1981) observed that an elevation in the levels of thyroxine exists in women during pregnancy. Reimers et al. (1984) examined this occurrence by evaluating the effects of reproductive state on stimulated concentrations of thyroxine in Beagle dogs. They found that diestrous and pregnant bitches had greater concentrations of thyroxine when compared to dogs in other

reproductive states. This suggests that reproductive status affects relative proportions of thyroxine secretion by the thyroid gland. Additionally, thyroxine levels can be affected by meals which contain moderate amounts of carbohydrates. Glade and Reimers (1985) discovered that serum thryoxine concentrations increased during the first several hours after meal completion in young horses. This increase in thyroxine after the consumption of a meal has also been identified in pigs (Ingram and Evans, 1980), humans (Danforth et al., 1979) and cattle (Blum et al., 1979). No differences in T_4 concentrations have been reported in weanling horses fed restricted diets, whereas in other species, reduced intakes decreased thyroid hormone concentrations (Buonomo and Baile, 1991).

Wang et al. (1980) evaluated the anterior pituitary of rats after thyroidectomy and ovariectomy and administration of subsequent injections of thyroxine. The anterior pituitary was removed, quartered, and placed in incubation flasks where they were rinsed to collect medium for assay of LH concentration. Rats that had their ovaries and thyroids removed had significantly higher serum LH and a greater response to GnRH, evident by a greater release of LH from the anterior pituitary, than did the ovariectomized rats. Thyroxine replacement depressed serum LH significantly. Overall, these data demonstrate that thyroidectomy enhances and thyroxine administration inhibits the synthesis and subsequent release of LH by the anterior pituitary of ovariectomized rats.

Insulin-like Growth Factor-1

Insulin-like growth factor-1 is a protein growth factor that under normal physiological conditions is primarily regulated by growth hormone (Buonomo and Baile, 1991). Tannenbaum et al. (1983) studied IGF-1 and its association with GH in rats and found that insulin like growth factors, which includes IGF-1, participated in a GH negative feedback at the level of the brain. The authors also suggested that the IGF family plays a role in maintenance of body weight and nutritional homeostasis at the central nervous system level. Chung et al. (1985) studied the association of GH and IGF-1 and found when GH was administered to swine that an elevation in IGF-1 concentrations occurred. In protein deficient mares Sticker et al. (1995) reported an increase in GH and a reduction in plasma IGF-1 concentration which is similar to reports in heifers (Houseknecht et al., 1988) and rats (Prewitt et al., 1982). Also, Buonomo and Baile (1991) studied the effects of feed restriction and subsequent refeeding on plasma levels of GH and IGF-1 in swine. Despite an elevation in plasma GH levels after 48 hr of feed restriction, circulating levels of IGF-1 levels were decreased by 53%. Refeeding fasted pigs was associated with a decline in GH but an increase was found in IGF-1 concentrations. Therefore, the authors concluded the GH-IGF-1 axis becomes uncoupled during nutritional restriction. Similarly, Vandehaar et al. (1995) studied heifers in a negative or positive energy balance and concluded those heifers fed below maintenance had smaller CL than did control heifers. The negative energy balance lowered levels of IGF-1 in serum but not in luteal tissue. The results also suggest that a negative energy balance "uncouples" the link between GH and IGF-1

In mammalian species, IGF's might play a key role in sensitizing ovarian granulosa cells to FSH action during terminal follicular growth as they are stimulators of follicular growth and maturation (Mazerbourg et al., 2003). IGF-1 is synthesized within the ovary and is found to stimulate the synthesis of progesterone (McArdle and Holtorf, 1989) by acting synergistically with gonadotropins to promote growth and steroidogenesis of ovarian cells (Lucy, 2000). IGF-1 may serve as an amplifier of gonadotropin hormonal action at the level of the granulosa cell (Adashi, 1998). A positive correlation between LH pulsatility and ovarian follicular development has been established for postpartum cows. Furthermore, a correlation between plasma estradiol during the first postpartum follicular wave and serum IGF-1 has been found in dairy cows (Beam and Butler, 1999).

The level of fatness an animal possesses will alter the serum IGF-1 concentrations. It has been shown that cows calving at a BCS of ≥ 5 have higher serum IGF-I concentrations postpartum than cows that calve in a BCS of <5. Furthermore, cows maintaining postpartum body condition have higher IGF-I levels compared to cows which lose body condition postpartum (Spicer et al., 2002). Comparatively, Oliver et al. (1993) stated that maternal starvation lowers both fetal nutrient and IGF-1 concentrations in sheep.

CHAPTER III

MATERIALS AND METHODS

Twenty-four pregnant Quarter Horse mares from the broodmare band belonging to the Texas A&M University Department of Animal Science were utilized for this study and equally divided into 2 treatment groups. Horses used in this study were maintained under the approval of the Texas A&M University Institutional Agricultural Animal Care and Use Committee using guidelines set forth by the Federation of Animal Science Societies (1999). The mares ranged in age from 3 to 18 y with an average age of 9 y, and a parity of 0 to 11 foals with the average number of foals being 4 foals per mare. Mares were assessed for body condition during the last week of October 2003 by 3 individual appraisers and assignment to groups was designated based on this evaluation. Groups were designed to contain mares with a desired body condition at least 1 month prior to foaling and subsequently monitored for maintenance of body condition up to time of rebreeding. Body condition scores were assigned on a scale from 1 to 9, including half-point increments, with a score of 1 representing an emaciated animal and a score of 9 representing an obese individual (Henneke et al., 1983). The 2 treatment groups were: 1) fat-conditioned mares (BCS range of 7 to 8) and 2) moderately-conditioned mares (BCS range of 5 to 6). All mares were blocked by expected foaling dates so that a proportionate number of mares in each treatment group foaled in the months of January through April.

Approximately 60 days before the birth of the first foal, the treatment groups were moved to separate pastures and fed to maintain the desired body condition. Each

group of mares was fed twice per day, with approximately 12 h between feedings. All

mares in each treatment group were fed a 13% crude protein, pelleted concentrate

(Appendix Table 30A) balanced accordingly for mares in late gestation and early

lactation (NRC, 1989). The amount of concentrate per day varied between the groups in

order to achieve desired body condition. Good quality bermudagrass hay along with

pasture grazing and water were available ad libitum. Concentrate intake was adjusted as

necessary to maintain the mares' body condition following foaling. Upon parturition,

the mares and foals were pastured with other mares and foals in the same treatment

group.

Physical Measurements

Mare's weight, body condition score and body fat percentage were determined

once every 2 wk beginning in December of 2003. Condition scores were assigned to the

mares by at least 2 independent appraisers. Rump fat thickness was measured 5.08 cm

from the midline and 10.16 cm from the point of the hip by ultrasonic scanning

equipment with a 5 MHz transducer (Medison SonoVet 600[®], Universal, Bedford Hills,

NY). Body fat content was estimated by the method of Westervelt et al. (1976). All

mares were assigned condition scores, weighed and fat scanned on the same day.

Additionally, at parturition each foal was measured for body weight, body length,

and wither height in order to calculate foal birth size.

Serum Sampling

Blood samples were collected daily by jugular venipuncture from the time of foaling until the detection of the second postpartum ovulation. The collections were accomplished between 10 a.m. and 2 p.m. each day to standardize comparisons of serum concentrations between feedings and to minimize hormone fluctuations due to feed consumption. Approximately 27 ml of blood was collected in evacuated venipuncture tubes. Following collection, the blood samples were allowed to clot for approximately 30 min at room temperature and then refrigerated at 4°C until centrifuged. The harvested blood was centrifuged in a refrigerated centrifuge for 45 min at 3000 x G. Following centrifugation, the harvested serum was transferred to microcentrifuge tubes and stored at -20°C until time of assay.

Estrous Cycle and Breeding

Beginning on approximately d 5 after foaling, mares were teased with a vigorous stallion in groups or individually to detect estrus. Follicular development was monitored by transrectal ultrasonography (Medison SonoVet 600®, Universal Medical Systems, Inc.-Bedford Hills, NY) every other day at the onset of behavioral estrus or 7 days post-foaling if no behavioral signs of estrus had been observed. Mares were not bred during the first postpartum estrus but were bred during the second postpartum estrus. Mares were artificially inseminated with extended semen when a follicle of 35 mm or greater was detected. An insemination dose of 500×10^6 progressively motile spermatozoa, extended in a ratio of 1:1 extender (E-Z Mixin®, CST-ARS, Chino, CA) to semen, was

placed into the mare's uterus. Insemination was repeated every other day until the

detection of ovulation of the dominant follicle. Transrectal ultrasonography was used to

determine pregnancy on approximately d 14 and 25 following ovulation. All mares were

maintained in their treatment groups until determined pregnant at 25 days post ovulation.

Radioimmunoassay (RIA) Procedures

Concentrations of LH (Williams et al., 1982), progesterone (Johnson et al.,

2003), leptin (Fitzgerald and McManus, 2000), thyroxin (Reimers et al., 1984), and IGF-

1 (Sticker et al., 1995) were analyzed by RIA as previously described for horse samples.

Progesterone, leptin, thyroxine, and IGF-1 assays were analyzed using a Packard Cobra

II® gamma counter. The LH assays were analyzed using a Packard Autogamma 5780®

counter. RIA kits contained all required reagents for completing the assay. All samples

from a given animal were assayed in a single assay for each hormone. Also, all samples

were run in duplicate and standard and reagents were run in triplicate.

Leptin was measured by a multispecies leptin RIA kit (Linco Research, Inc., St.

Charles, MO). A sample volume of 100 µl was used with a sensitivity of 1.0 ng/ml. The

intra- and inter-assay coefficient of variation between the 2 controls was 2 and 11% and

7 and 9% (n = 6 assays), respectively.

Serum thyroxin and progesterone were analyzed using single-antibody RIA kits.

(Coat-A-Count®, Diagnostic Products Corp., Los Angeles, CA). The kits contained all

required reagents including antibody-coated polypropylene tubes, iodinated thyroxin and

progesterone, and standards. A sample volume of 100 µl was used for each assay with a

sensitivity of the progesterone assay equaling 0.1 ng/ml and the thyroxine assay equaling

1.0 µg/dl. The intra- and inter-assay coefficients of variation for thyroxin were 5 and

13% (n = 4 assays), respectively. The intra- and inter-assay coefficients of variation for

progesterone were 6 and 3% (n = 5 assays), respectively.

Serum IGF-1 concentrations were measured by an RIA kit (Diagnostic Systems

Laboratories, Inc., Webster, TX). A sample volume of 50 µl was used with a sensitivity

of 0.8 ng/µl. The intra- and inter-assay coefficient of variation between the controls was

3, 8 and 18% (n = 5 assays), respectively.

LH concentrations were measured by an RIA over a 4-day period. On d 1, 500

µL of 1% phosphate buffered saline (PBS) with egg white (PBS-EW) was added to the

non-specific binding (NSB) and the 0 standard tubes. Two-hundred microliters of

standard and 300 µl of 1% PBS-EW were added to each standard tube. Three-hundred

microliters of 1% PBS-EW along with 200 µl of each sample were put into each

unknown tube. The reference preparation tubes contained 300 µl of 1% PBS-EW and

200 µl of reference preparation. The primary antibody was anti-eLH, which was diluted

with PBS-EDTA and normal rabbit serum (NRS) in a 1:400 ratio. Two-hundred

microliters of PBS-EDTA and NRS without the primary antibody was added to the NSB

tubes only. Two-hundred microliters of the antibody was then added into all tubes with

the exception of the NSB and total count tubes. A tracer consisting of 100 µl of ^{125}I-eLH

(20,000 CPM/100 µl diluted in 0.1% PBS-EW) was added to all tubes and then vortexed

and allowed to incubate for 24 h at 4°C. On d 2, 200 µl of sheep-anti-rabbit gamma

globulin diluted in PBS-EDTA without NRS was added to all tubes except the total

count. Tubes were once again incubated at 4°C for 48 to 72 h. On d 4, 3.0 ml of ice cold PBS (0.01 M; pH 7.0) was added to all tubes except for the total count. The samples and reagents were then centrifuged at 3600 RPM for 1 h while maintained at 4°C. Once centrifugation was complete the tubes were decanted and supernatant discarded. Tubes were then counted in a gamma counter. The intra- and inter-assay coefficients of variation for the controls for LH were 15% and between 5 and 20% (n = 2 assays), respectively.

Statistical Analysis

Body condition scores, rump fat thickness, and percentage body fat, along with foal birth weights were analyzed by 2-way ANOVA using Stata Statistical® software version 8.0 (Stata Corporation, College Station, TX). This method was also used to analyze the mean number of days from parturition to the first ovulation, second ovulation, and interovulatory interval. Group, time, and group x time interactions were analyzed. Additionally, serum concentrations of each hormone were standardized for comparison during the estrous cycle and analyzed by analysis of variance for mean concentration from parturition to first ovulation, parturition to second ovulation, and the interovulatory period between the 2 treatment groups. Group, time, and group x time interactions were also analyzed. All data were considered significant if probability was less than 0.05.

CHAPTER IV

RESULTS

Body Condition

Mares were evaluated for body condition to insure the correct body fatness level for the group to which they were assigned and to insure there was a difference between the 2 groups. Semi-monthly analysis of BCS, rump fat thickness and percentage body fat revealed that the fat group was significantly higher in these 3 areas (P<0.0001). As the mares approached time of parturition, mares in the fat group achieved a desired BCS of 7.17 and those in the moderate group achieved a BCS of 5.43 (Table 1). The greater thickness in rump fat and elevated percentage body fat further substantiate the differences between the 2 groups of mares. As was expected, body condition, rump fat thickness and percentage body fat all declined following parturition. Nevertheless, the desired BCS was maintained for each group as the fat mares had a mean BCS of 6.98 and moderate mares had a mean BCS of 5.01. This higher body condition was also evident by a thicker rump fat covering in the fat mares which correlates into a higher body fat percentage. The data indicate that each group of mares was in the desired body condition score range to meet the requirements of this study, both before and after parturition (Table 1).

Although mares in each group maintained the desired body condition both before and after parturition, moderate conditioned mares had a greater loss in percentage body fat when compared to fat conditioned mares upon foaling. This was determined by the difference in percent body fat from 1 to 10 days prior to foaling versus 1 d after foaling.

Mares in the fat group lost 0.35 % body fat, whereas the moderate mares lost 0.82 %

body fat. Treatment group did not affect the change in mare weight (Table 2).

Table 1. Mean (±SE) body condition score, rump fat thickness, and percent body fat of fat- vs moderate-conditioned mares before and after parturition

Group	n	BCS	Rump Fat (cm)	%Body Fat
Fat				
Before:	12	7.17 ± 0.06^a	1.33 ± 0.06^a	14.90 ± 0.28^a
After:	12	6.98 ± 0.03^a	1.14 ± 0.05^a	14.00 ± 0.23^a
Moderate				
Before:	12	5.43 ± 0.06^b	0.96 ± 0.06^b	13.18 ± 0.26^b
After:	12	5.01 ± 0.07^b	0.70 ± 0.04^b	11.97 ± 0.20^b

[a,b] Means within column lacking common superscripts differ (P<0.0001).

Table 2. Mean (±SE) percentage change in body fat and change in mare weight measured within 1 wk prior to foaling versus 1 d post foaling

Group	n	Change in %Body Fat	Change in wt (kg)
Fat	12	$- 0.35\pm0.17^a$	-69.55 ± 5.16^a
Moderate	12	$- 0.82\pm0.12^b$	-76.35 ± 5.83^a
Mean Total	24	$- 0.58\pm0.11$	-72.95 ± 3.87

[a,b] Means within column lacking common superscripts differ (P<0.05).

Days to Ovulation

As indicated in Table 3, the time from parturition to the first postpartum ovulation, or foal heat ovulation (Ovulation 1), for mares in the fat group was 14.41 days and for mares in moderate body condition was 16.18 days (P>0.05). The time from foal heat ovulation to the second postpartum ovulation, or interovulatory period (IOP), was 22.91 days for mares in a fatter body condition and 24.33 days for moderately conditioned mares (P>0.05).

Table 3. Mean (±SE) interval from foaling to first ovulation (Ovulation 1) and days from first to second ovulation, or interovulatory period (IOP), of fat- vs moderate-conditioned mares

Group	n	Ovulation 1 (d)	IOP (d)
Fat	12	14.41±1.07	22.91±1.07
Moderate	12	16.18±1.06	24.33±0.93
Total	24	15.26±0.77	23.65±0.71

Means within column are not different (P>0.05)

Rebreeding Efficiency

The rebreeding efficiency of the mares in each group was determined by rectal ultrasonography for detection of an embryo on d 14 after ovulation during the second postpartum estrus. The pregnancy rates were similar (P>0.05) between the fat- and moderately-conditioned mares at 91.67 and 83.33%, respectively (Table 4). There were no twin ovulations detected from any mares throughout the study.

Table 4. Number and percentage of mares confirmed pregnant by rectal ultrasonography at d 14 post-ovulation

Group	n	Pregnant at d 14	% Pregnant
Fat	12	11	91.67
Moderate	12	10	83.33
Total	24	21	87.50

Percentage pregnant within column are not different (P>0.05)

Foal Measurements

Mean foal birth weight, height, and body length for mares in either a fat- or moderate- body condition are included in Table 5. The foals of the fat mare group averaged 47.74 kg for weight, 98.96 cm for height and 74.74 cm for length. Comparatively, the foals of the moderate mare group weighed 50.24 kg, and had heights and lengths of 100.86 cm and 74.10 cm, respectively. The measurements were not different between the groups (P>0.05).

Table 5. Mean (±SE) weight (Wt), height (Ht), and length (Lg) of foals measured on day of birth from fat and moderate conditioned mares

Mare group	Wt (kg)	Ht (cm)	Lg (cm)
Fat	47.74±1.54	98.96±0.94	74.73±1.04
Moderate	50.24±2.59	100.86±1.32	74.10±.1.24
Total	48.99±1.50	99.90±0.81	74.40±0.79

Means within column are not different (P>0.05)

Hormone Analyses

Serum LH and Progesterone. Analyses of serum LH following parturition indicate that rectal ultrasonography was accurate in determination of time of ovulation as Figure 1 illustrates the pattern of LH concentration during the estrous cycle for the fat- and moderate-conditioned mares, respectively. Mean serum concentration of LH increased prior to the first ovulation, remained elevated until approximately d 2 post ovulation and declined by d 5 (Figure 2). Mean serum LH concentrations did not differ between the groups at the first postpartum ovulation (P>0.05). Mean LH concentration was lower P<0.05) in the fat- than in the moderate-conditioned group during the period from d 13 to the time of second ovulation (Figure 3). Mean LH concentration for each sampling day from d-13 to time of ovulation was reduced (P<0.0001) in the fat-compared with the moderate-conditioned mare group.
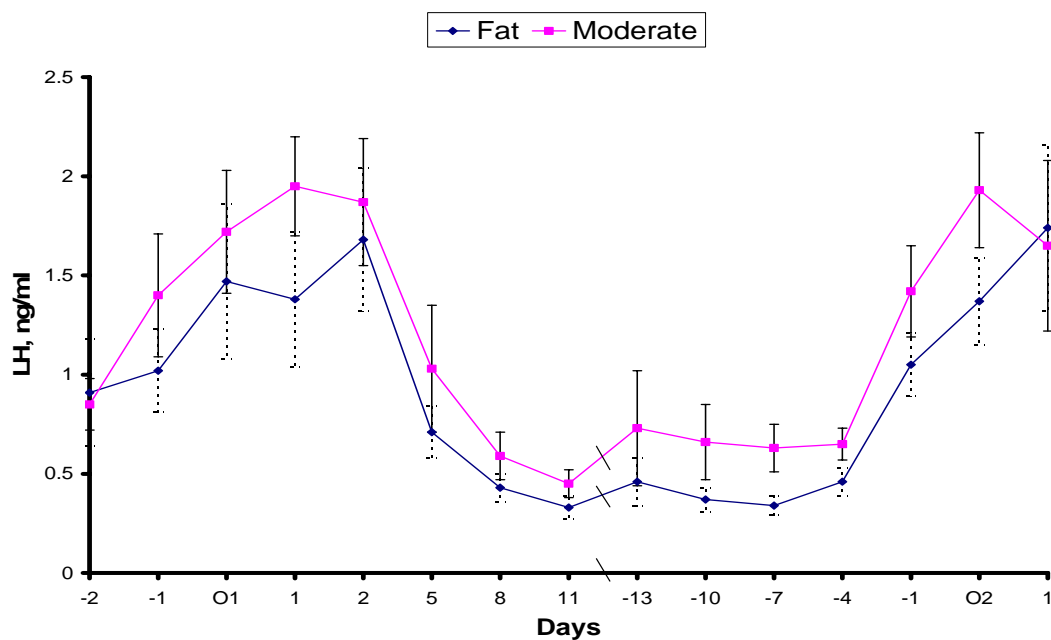
Figure 1. Mean serum concentrations of LH in fat- and moderate-conditioned mares during the estrous cycle following parturition (O1 = first ovulation; O2 = second ovulation).
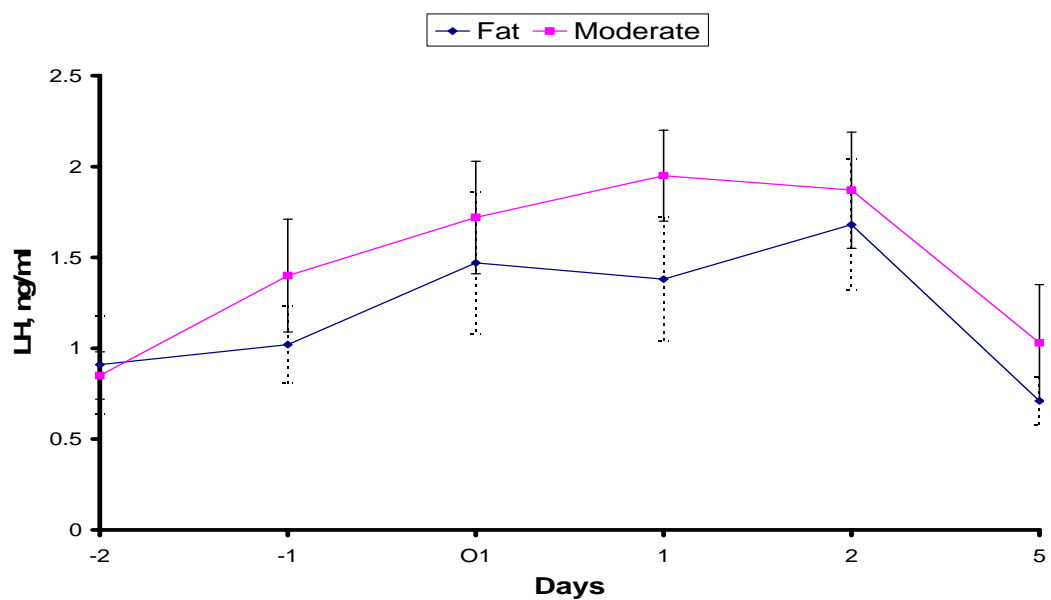

Figure 2. Mean serum concentrations of LH in fat- and moderate-conditioned mares around time of first ovulation post foaling (O1 = first ovulation).
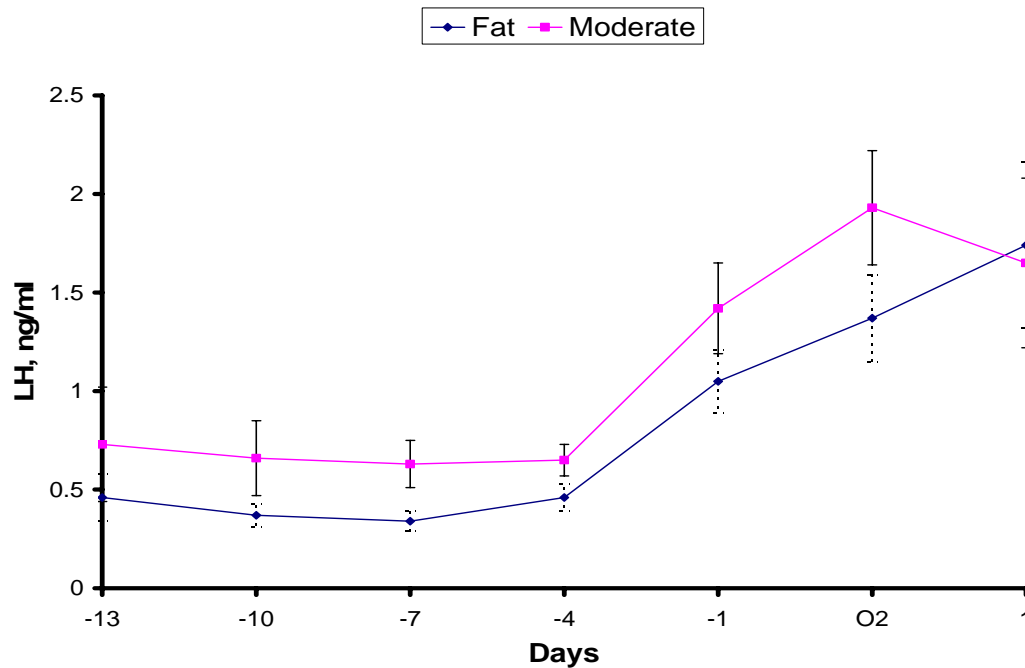
Figure 3. Mean serum concentrations of LH in fat- and moderate-conditioned mares approaching the second postpartum ovulation (O2 = second ovulation).

The pattern of serum progesterone concentrations indicates that ultrasonogram detection of ovulation was accurate (Figure 4). Mean serum progesterone concentrations were not different for fat- versus moderately-conditioned mares as measured from ovulation 1 to ovulation 2. Peak progesterone concentrations around d 6 post ovulation indicate that ovulation did occur, and was followed by the development of a functional CL. Mean progesterone concentrations did not differ (P>0.05) between groups during the interval between the first and second postpartum ovulations.

Figure 4. Mean serum concentrations of progesterone in fat- and moderate-conditioned mares during the estrous cycle following parturition (O1 = first ovulation; O2 = second ovulation).

Serum Leptin. The mean serum concentrations of leptin did not differ (P>0.05) between the 2 groups of mares at any time during the postpartum days to first ovulation, from first to second ovulation or on the days during the interovulatory period (Figure 5). Mean leptin concentration from 3 days prior to 3 days after the first ovulation was 2.38 ± 0.26 ng/ml. Mean leptin concentration during the interovulatory period was 2.20 ± 0.08 ng/ml. Mean leptin concentration during the second postpartum ovulation, 2.10 ± 0.17 ng/ml, consisting of 3 days before ovulation to the day of ovulation, was not different (P>0.05) as there was no group x day interaction detected.

Figure 5. Mean serum concentrations of leptin in fat- and moderate-conditioned mares during the estrous cycle following parturition (O1=first ovulation; O2=second ovulation).

Serum Thyroxine. Serum T_4 concentrations were significantly different between the 2 groups throughout the estrous cycle following parturition. Figure 6 shows that during the interval from 3 days before to 3 days after the first ovulation ($P<0.01$), during the interovulatory period ($P<0.01$), and the period 3 days before and including the second ovulation ($P<0.0001$), the mean T_4 concentrations were higher in mares of a moderate body condition compared with those of a fatter body condition.
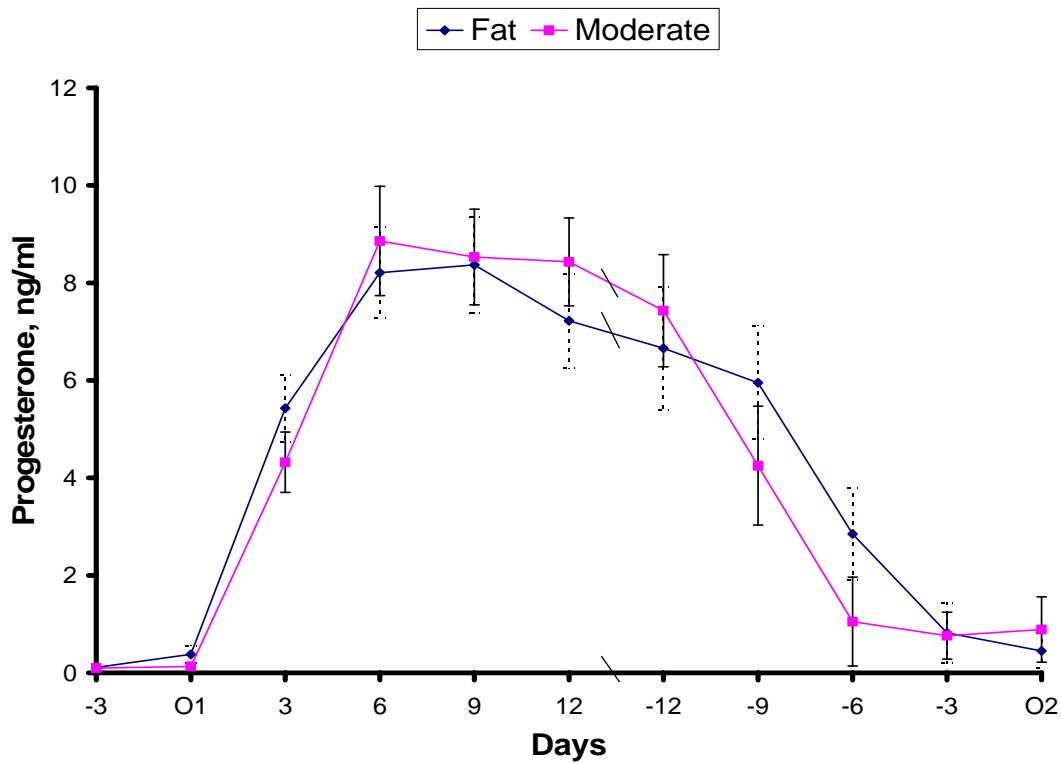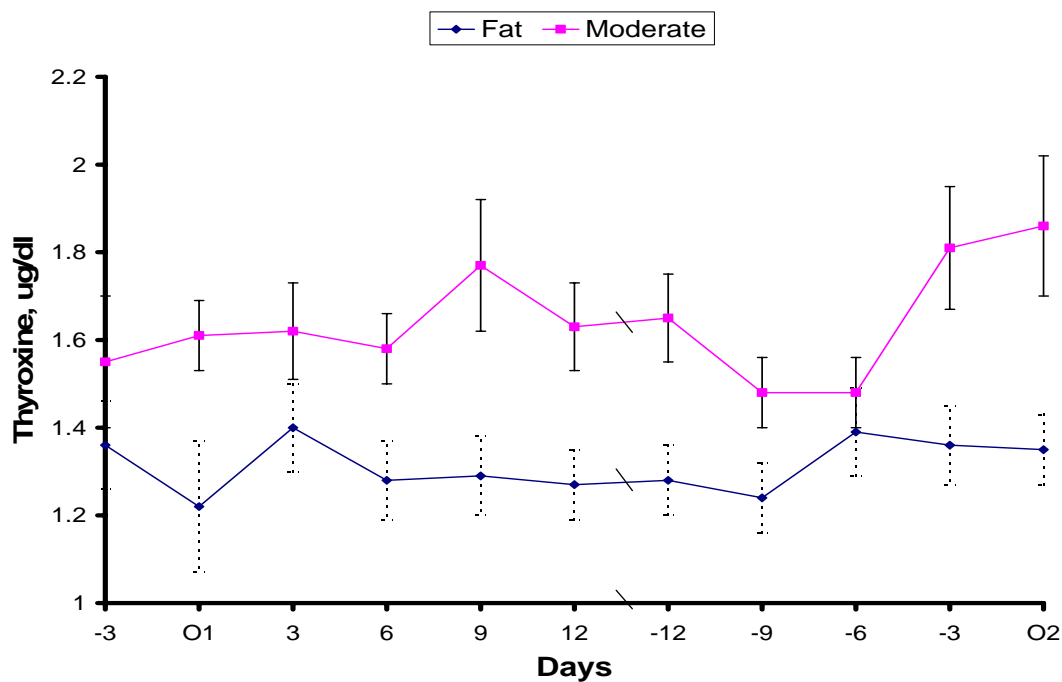
Figure 6. Mean serum concentrations of thyroxine in fat- and moderate-conditioned mares during the estrous cycle following parturition (O1 = first ovulation; O2 = second ovulation).

Serum IGF-I. Mean serum IGF-1 concentrations between the 2 groups were significantly different throughout the estrous cycle (Figure 7). In the days around the first postpartum ovulation (Figure 8), mares in a fat body condition had consistently higher concentrations of IGF-1 (P<0.01). Furthermore, as the mares progressed through the interovulatory period (P<0.01) and approached the second postpartum ovulation (P<0.01) mean serum IGF-1 concentrations remained elevated for the mares in the fat conditioned group (Figure 9).
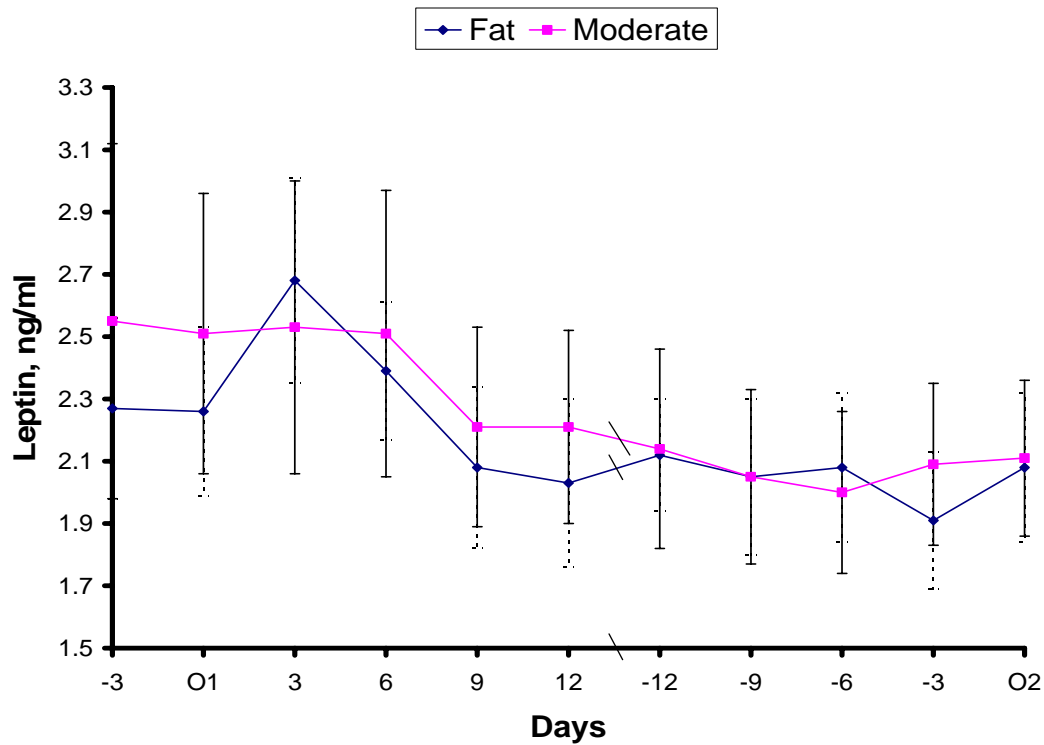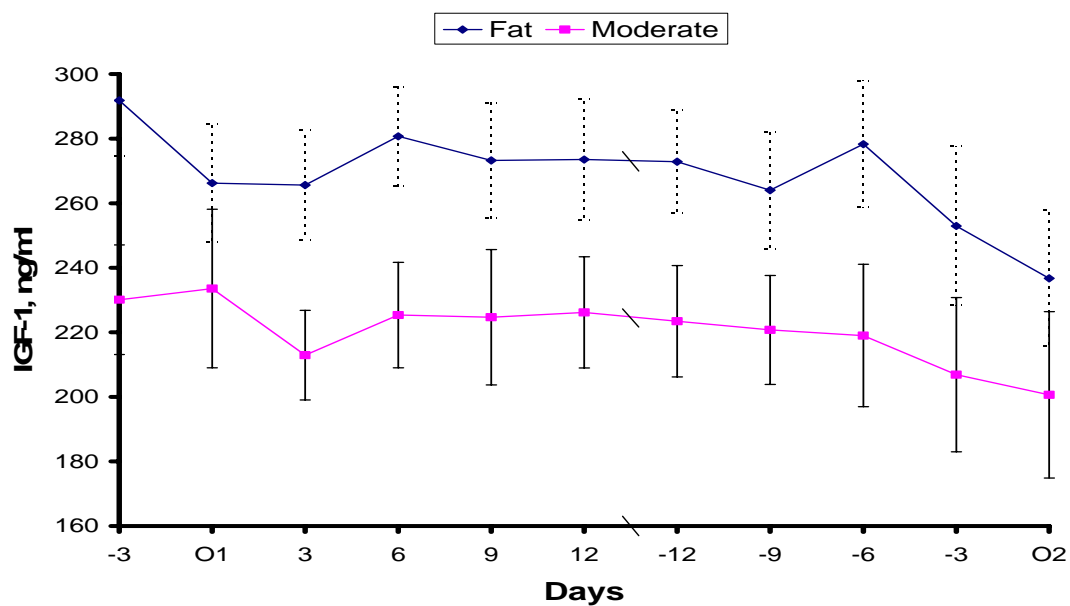
Figure 7. Mean serum concentrations of IGF-1 in fat- and moderate-conditioned mares during the estrous cycle following parturition (O1 = first ovulation; O2 = second ovulation).

Figure 8. Mean serum concentrations of IGF-1 in fat- and moderate-conditioned mares around time of first ovulation post foaling (O1).
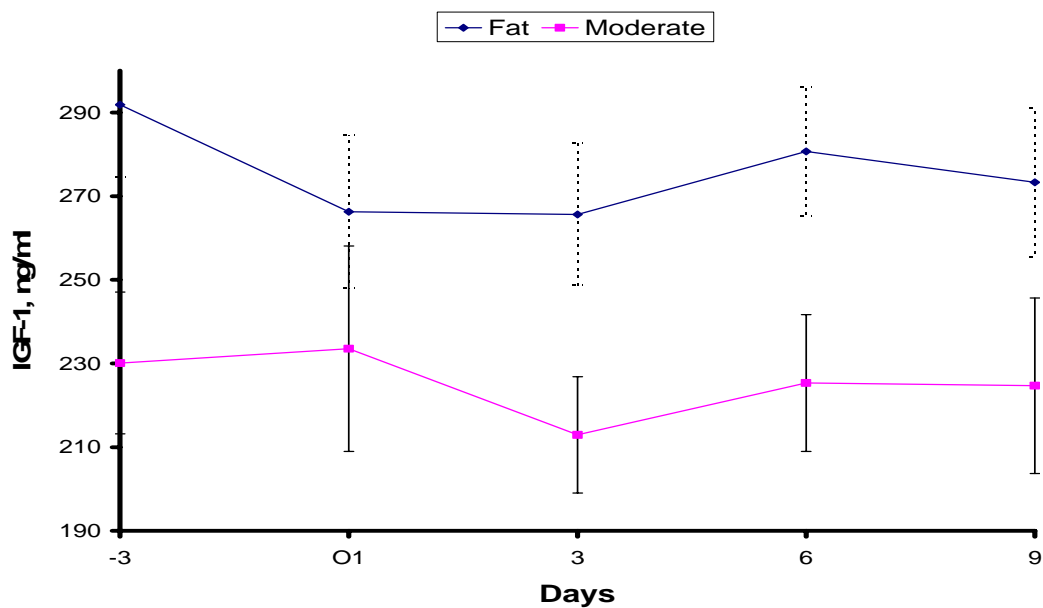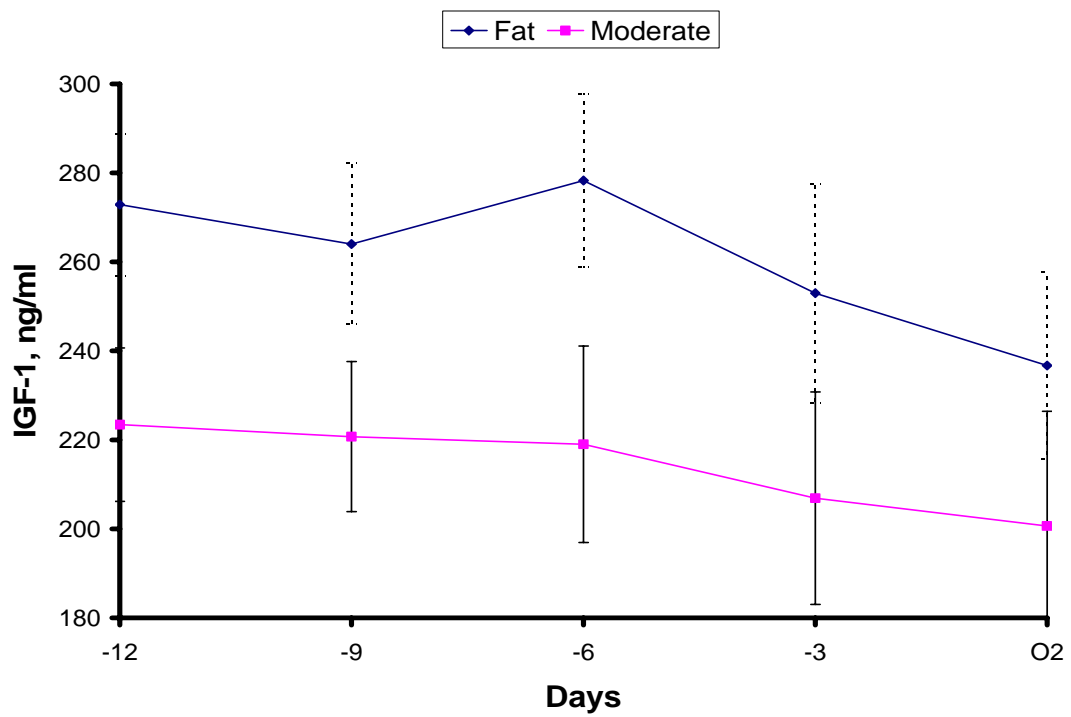
Figure 9. Mean serum concentrations of IGF-1 in fat- and moderate-conditioned mares approaching the second postpartum ovulation (O2).

CHAPTER V

DISCUSSION

Body Condition

The 2 groups of mares were maintained in the appropriate body condition throughout the study, as not only were the BCS consistently higher in the fat mare group but also the rump fat was thicker, which indicated a higher percentage body fat. This was evident both before and after parturition. After foaling, mares in both groups lost rump fat and possessed a lower percentage of body fat as was expected of mares in late gestation (Powell et al., 1989) and early lactation. A similar finding in cows suggests that energy restriction in late prepartum lowers body condition at calving (Whitman, 1975). Interestingly, the mean decrease in body fat percentage was higher in the moderate- as compared to the fat-conditioned mares from before foaling to after foaling. The differential response may have been due to the fat conditioned mares using less energy reserves during parturition and milk synthesis as their energy intake was higher throughout the study. The moderate mares were unable to increase energy intake enough to meet the needs of fetal growth and lactation and therefore mobilized body fat to meet these requirements instead. The rapid decrease in body fat resulted in a lower BCS for all mares shortly after parturition; however the change was more drastic in mares maintained in a lower body condition. This lowered body condition will cause the mare foaling in a BCS of a 5 to possibly drop as low as a BCS 4. Prior research has shown that females in a BCS of less than 5 may experience reproductive inefficiencies (Henneke et al., 1981; Richards et al., 1986; Hines et al., 1987; Lake et al., 2005) which

will leave the female at a reproductive disadvantage after foaling as compared to those that achieve and maintain a fleshier body condition.

The statement has been made that prepartum and postpartum energy balance are the most important factors affecting duration of the postpartum interval to estrus in beef cows (Hess et al., 2005). Previous work has indicated that mares in a BCS of less than 5 will be more likely to have longer interovulatory periods, lower pregnancy rates at 30 days post-ovulation, lower pregnancy maintenance to 90 days (Henneke et al., 1984), and longer mean gestation lengths (Hines et al., 1987). Studies in cattle show that cows in a BCS of less than or equal to 4 as compared to greater than or equal to 5 have lowered pregnancy rates by up to as much as 30% (Rae et al., 1993) and if not lactating, can undergo anestrus (Richards et al., 1989). However, it is important to determine the threshold BCS for optimum rebreeding efficiency of mares and to also determine if any problems exist with mares being overly conditioned. As the results of this study indicate, there was no evident difference in reproductive efficiency between the 2 different groups of mares as the mean number of days to first cycle ovulation (14.41 ± 1.07 vs 16.18 ± 1.06), mean number of days from first to second cycle ovulation (22.91 ± 1.07 vs 24.33 ± 0.93), and mean conception rates (91.67% vs 83.33%) at 14 days post-ovulation were not different (P>0.05). The mean days to ovulations are very similar to the findings of Henneke et al. (1984) and suggest that mares of a moderate body condition will achieve as short of a period from parturition to ovulation as mares maintained in a fleshier body condition and will also have a similar length of time between ovulations after foaling. This is in agreement with Kubiak et al. (1989) who

found that the interval from foaling to first cycle ovulation, foaling to second cycle ovulation, and first to second cycle ovulations were similar in mares held in a BCS of 9 versus those mares in BCS of 5.5 to 7. The current research further expands the range of adiposity presented by Kubiak et al. (1989) that will achieve reproductive efficiency in the mare. In contrast, it does not agree with the findings of Fitzgerald et al. (2003) who concluded that mares of a BCS of 7 or higher had lengthened luteal phases and enhanced length of interovulatory periods as compared to lean mares in a BCS of 4. However, in the study by Fitzgerald et al. (2003) only 12 total mares ranging in age of 15 yr or higher were used, and the time period analyzed encompassed both natural breeding and non-breeding seasons (July thru February). Previous reports have confirmed that aged mares (≥15 yr) have increased endometrial inflammation, reduced pregnancy rates, increased embryo loss rates (Carnevale and Ginther, 1992) and some aged mares may possess ovaries that have reached senescence (Ginther, 1992) which all lead to lowered fertility. Also, the time of year of the natural breeding season of the mare is the spring and summer months. During the fall and winter months the incidence of ovulation is minimal or absent (Ginther, 1992) as concentrations of reproductive hormones in serum and pituitaries of horses are highest during the breeding season and lowest during the nonbreeding season. These protocols may be contributing factors that give inaccurate data and lead to conflicting reports that thin mares are more reproductively efficient than fat mares following parturition.

The current results give a more accurate assessment as to the range of adiposity that is acceptable to enhance reproductive capabilities of the foaling/lactating mare.

DeRouen et al. (1994) states that cows calving in a BCS of 6 to 7 have higher pregnancy rates compared to those with BCS of 4 to 5 with the interval from parturition to pregnancy for cows calving at a low BCS being 10 to 18 days longer than for those of a higher BCS. Mares in both groups achieved and maintained pregnancy, further indicating the similarity of reproductive efficiency between the groups when fed to achieve and maintain a BCS of at least a 5. Results of the current study indicate that mares should be maintained in a BCS of 5 or higher following parturition through the second postpartum ovulation in order to achieve the shortest intervals from foaling to ovulation, maintain a normal interovulatory interval, and achieve optimum conception rates. However, following parturition mares will lose body fat; therefore, it is vital that the mare be in a body condition score of at least 6 during this critical time in order to maximize the rebreeding efficiency of the postpartum, lactating mare. This is important for the mare breeder or owner in order to enhance the reproductive capabilities of the mare while at the same time minimizing maintenance cost of the broodmare.

Rebreeding efficiency was not different between the groups. However, the fat mare group did achieve a numerically higher percent conception rate versus the moderately conditioned mares. This was somewhat similar to the findings of Henneke et al. (1984) who observed that mares maintained in a low body condition prior to foaling experienced lower pregnancy rates versus mares in a higher body condition (50% vs 100%) after parturition. It is also in agreement with a previous study in beef cows indicating calorie restriction during gestation reduced pregnancy rates after calving (Wiltbank et al., 1962). Theoretically, there is a difference between 91.67% and 83.33%

to the mare breeder if the number of mares being bred is substantial. For someone who breeds 100 mares in a given breeding season, 91 foals versus 83 foals could result in significant economic and time loss to the breeder. However, further studies should be conducted, with a larger sample size, in order to be certain of a speculated difference in rebreeding efficiency between mares of a fat- versus moderate-conditioning.

Foal Measurements

It has been shown that extremely obese women have significantly heavier babies than thinner females (Ekblad and Grenman, 1992). Birth weight has been shown to be influenced by the BCS of the cow at time of parturition (Wiltbank et al., 1962; Anthony et al., 1986; Spitzer et al., 1995). Additionally, mare phenotype and genotype have been shown to influence the size of offspring due to the availability of area in the uterine body (Allen et al., 2002). However, the foals' mean birth measurements, as defined by body weight, body height, and body length, were not found to differ (P>0.05) between the 2 groups of mares. The mean measurements of foals born from fat- and moderate-conditioned mares were: body weight of 47.74 ± 1.54 kg versus 50.24 ± 2.59 kg, wither height of 98.96 ± 0.94 cm versus 100.86 ± 1.32 cm, and body length of 74.73 ± 1.04 cm versus 74.10 ± .1.24 cm, respectively. In cattle, conflicting reports have indicated that nutrient intake may either alter the birth weights of calves by increasing birth weights when cows were fed a high level of energy (Wiltbank et al., 1962), or simply may not affect birth weights (Wiley et al., 1991). Also, birth weights have been found to be correlated with body condition of the female as the occurrence of dystocia is less and

birth weights are greater in primiparous beef cows who are in a higher body condition

score versus cows in a lower body condition score (Spitzer et al., 1995). The similarity

in birth measurements of foals born to mares in both groups is in part due to the

consistency of breed type amongst all mares and may also be due to the closeness of

body condition between mares of fat and moderate conditioning further indicating

reproductive similarities of mares in this body condition range.

Differences in foal measurements may have been observed if the mares were in a

body condition score less than 5. At this point, they may not have had the energy

reserves required of carrying a foal to term, experienced a premature delivery or

delivered foals of less than normal weight as is found in studies with underweight

women (Brown et al., 1981).

LH and Progesterone

Serum concentrations of LH and progesterone were monitored in order to

correlate ultrasonography with the normal hormonal patterns of the mare during the

estrous cycle. Therefore, data such as times of ovulation and periods of non-estrus were

compared to actual LH and progesterone serum concentrations.

The values for serum LH concentrations in this study range from 0.33 ng/ml

during the interovulatory period to 1.95 ng/ml at the time of ovulation and are not

similar to the highest and lowest values given for serum LH of some texts such as

Ginther (1992). However, the absolute levels of LH in tabulated reports vary as much as

191-fold among studies. This is attributed to widely diverse assay procedures, especially regarding the purity of the LH standards (Ginther, 1992).

A high-frequency mode of pulsatile LH secretion is important for the final phase of maturation of ovarian follicles and thus for induction of estrus and ovulation (Schillo, 1992). In mares of both body condition groups, mean serum LH concentrations began to rise a few days before ovulation and were maintained a few days following ovulation before declining. During the interovulatory period, serum LH concentrations were at their lowest until once again rising as the second cycle ovulation approached. The cyclicity of serum LH of all mares in this study followed the expected pattern of a mare's estrous cycle according to Miller et al. (1980). The mean serum concentrations of LH were not different for mares of either group during the first cycle ovulation and the interovulatory period (P>0.05). Interestingly, mean serum concentrations of LH were higher (P<0.05) in mares of the moderate- versus fat-conditioned group during the days leading to the second cycle ovulation. In contrast, restriction of dietary energy late in the prepartum period or early in the postpartum period has been shown to reduce the number of animals returning to estrus within a well-defined breeding season. These effects are apparently associated with a failure to develop preovulatory follicles (Schillo, 1992) possibly due to the lack of LH or responsiveness of the ovary to LH. This effect was seen in a study conducted by Perry (1990) that found cows fed a low-energy diet before calving exhibited a lower frequency of LH pulses than cows that received a higher plane of nutrition. The moderately conditioned mares of this study must have consumed adequate dietary energy such that LH concentrations were not inhibited. The

fact that the moderate conditioned mares maintained high levels of LH, even higher than fatter-conditioned mares, is evidence that mares in a BCS of 5 to 6 have adequate LH in blood circulation to initiate ovulation and are therefore reproductively efficient following parturition. Even though the moderate conditioned mares had significantly higher serum LH concentrations during the days encompassing the second ovulation as compared to the fat conditioned mares, both groups ovulated within a normal time frame after foaling and the times to ovulation were not different between the 2 groups (P>0.05).

Additionally, serum concentrations of progesterone followed the expected pattern of the estrous cycle of the mare and no differences were detected in the serum of mares from either group (P>0.05). Progesterone concentrations were minimal at times of ovulation and reached a plateau approximately 6 days after ovulation giving evidence of a fully functional CL. Approximately 6 days prior to the second ovulation, progesterone concentrations fell drastically from their high levels during the interovulatory period. This described progesterone profile is in agreement with Towson et al. (1989).

The values of serum progesterone concentrations range from the lowest amount detectable, with the assay used, of 0.1 ng/ml during the time of ovulation to a high of 8.86 ng/ml during diestrus. These concentrations are similar to those reported by Ginther (1992). Progesterone concentrations in the blood do not increase until approximately d 3 postovulation in cattle and sheep (Ginther, 1992), however concentrations begin to rise within 12 to 24 hours following ovulation in mares (Plotka

et al., 1975). The current results also indicate that very soon after ovulation progesterone levels were measurably elevated.

The similarities between the LH and progesterone profiles of mares in both groups, along with the comparisons made in previous reports, conclude that the hormonal control of the estrous cycle of mares in this study was normal and fully functional.

Leptin

Leptin is characterized as a satiety hormone secreted from the adipocyte and blood concentrations have been shown to increase or decrease with the fluctuation of adiposity the animal possesses (Considine et al., 1996; Kolaczynski et al., 1996). Leptin may also play a role in reproduction as this hormone may be a signal to the female's body that she possesses an adequate amount of body fat to sustain a pregnancy (Barash et al., 1996; Bray, 1996; Houseknecht et al., 1998). If the female's fat stores reach a critical limit then there is an increased release of leptin into circulation. Leptin then acts upon hypothalamic cells to stimulate gonadotropin release (Yu et al., 1997) thereby having an influence on reproductive potential. There is evidence that a correlation exists between body condition and circulating concentrations of leptin, which shows that as body fat stores increase leptin levels also rise, and vice versa. The same effect of body fat mass on the amount of circulating leptin has been found in mares (Gentry et al., 2002b), cows (Lents et al., 2005), pigs (Bidwell et al., 1997), humans (Considine et al., 1996), and sheep (Blache et al., 2000). The influence of body fat on the concentration of

leptin further lends support to the fact that leptin is a signal as to the degree of fatness an animal possesses. This also may be another factor involved in the lowered reproductive capability of females with inadequate fat stores. In the current study, the analyses of mean serum concentrations of leptin between mares of fat- and moderate-conditioning revealed no differences at any time during the estrous cycle following parturition (P>0.05). This finding suggests that there is not enough difference between mares of a moderate or fat body condition to alter leptin concentrations during the estrous cycle. The likeness of leptin profiles between the 2 groups may be somewhat responsible for the similarities in reproductive efficiency. Additionally, the lack of a difference in mean leptin concentrations is evidence that mares in a BCS 5 or 6 have enough body fat to be reproductively sound and are equivalent to mares in a higher BCS such as 7 or 8.

Thyroxine

Mean serum T_4 concentrations were different between the 2 groups of mares throughout the duration of the estrous cycle following parturition (P<0.01). Thyroxine has been shown to exert an influence on the transition from the breeding season to anestrus in ewes (Karsch et al., 1995). If the thyroid is extracted from the animal the transition from the breeding season to anestrus is blocked and the female continues with normal estrous cycles (Moenter et al., 1991; Webster et al., 1991; Karsch et al., 1995). Apparently, T_4 has a suppressive effect on LH pulsatility but only during the transition into anestrus and not during the normal breeding season (Moenter et al., 1991) and the determination has been made that thyroxine concentrations do not fluctuate during the

estrous cycle (Johnson, 1986). However, another report postulates no change in T_4 concentrations during the estrous cycle except a decline following ovulation (Kelley et al., 1974). This was not the finding in the current study as there was not a decline in thyroxine concentrations from before to after ovulation. The mares of the current study were not monitored during the transition into the anestrus season, so the effect of T_4 for seasonal transition cannot be evaluated in mares in this study.

The reason for the lower mean concentration of T_4 in the fat conditioned mares may be due to the higher energy intake from an increased volume of concentrate consumption versus the intake of the moderate conditioned mares. Previous research has demonstrated conflicting reports on the effect of feed intake on thyroxine concentrations. Reports indicate that increased energy intake increases T_4 concentrations (Glade and Reimers, 1985), lowers T_4 concentrations (Buonomo and Baile, 1991), or simply has no effect (Sticker et al., 1995). The current study supports the claim that mares maintained in a higher body condition will have higher levels of thyroxine throughout the estrous cycle as compared to mares maintained in a lower body condition.

Insulin-like Growth Factor-1

Insulin-like growth factor-1 plays an important role in the maintenance of body weight (Tannenbaum et al., 1983) and this correlation was apparent as the serum concentrations of IGF-1 were shown to be higher in fat- as compared to moderate-conditioned mares in this study ($P<0.01$). The determination that fat mares possess greater concentrations of IGF-1 in the serum is in agreement with both Sticker et al.

(1995) and Spicer et al. (2002) and is consistent with studies in heifers (Houseknecht et al, 1988) and rats (Prewitt et al., 1982).

Insulin-like growth factor-1 may serve as an amplifier of gonadotropin hormonal action at the level of the granulose cell (Adashi, 1998; Lucy, 2000). The IGF system has also been postulated to have an intrafollicular role leading to follicle selection and ovulation (Ginther et al., 2004). If this is true, then greater concentrations of IGF-1 would correspond with greater LH and FSH concentrations and lead to follicle stimulation and ovulation. In the current study, LH concentrations did not differ between groups of mares throughout the estrous cycle ($P>0.05$) except during the approach to the second postpartum ovulation ($P<0.05$). At this time, the moderate conditioned mares actually had significantly higher concentrations of LH than did the fat mares while at the same time not having significantly different IGF-1 concentrations. In the current study, IGF-1 concentrations may have been sufficient in both groups of mares to elicit normal ovulation as the days to ovulation, along with conception rates, indicate equal reproductive efficiency. Insufficient levels of IGF-1 may be common in mares of a body condition below 5 as heifers maintained in a negative energy balance resulted in decreased IGF-1 concentrations and smaller CL when compared to controls (Vandehaar et al., 1995).

CHAPTER VI

SUMMARY

In this study, the analyses of reproductive efficiency between mares of a fat- or moderate-body condition suggests that the period of time to ovulation after foaling, the interovulatory period, along with foal measurements at birth are not different. The data show that mares maintained in a BCS 5 or higher have very similar reproductive capabilities following parturition. Furthermore, data provided in this study provide an array of measures of adiposity (BCS ≥ 5, rump fat ≥ 0.70 cm and body fat $\geq 11.97\%$) that are associated with efficient rebreeding in the lactating mare. However, it is important to note that moderate conditioned mares will lose more body fat following parturition than will fleshier conditioned mares. Therefore, it is ideal to have the gestating broodmare in a BCS of 6 or higher as she approaches time of parturition in order to insure maximum rebreeding efficiency after foaling.

Leptin is a signal to the brain of the amount of fat the body possesses. This signal is hypothesized to be an indicator to the animal of whether or not it has the energy reserves to support reproduction. Serum leptin levels remained similar in both groups of mares and the similarity between the groups indicate that the moderate conditioned mares possessed adequate amounts of fat for reproductive performance. The higher concentrations of T_4 found in the moderate conditioned mares is likely due to the intake of a lower volume of concentrate which correlates into lower energy intake and a lower percentage of body fat. Previous reports have shown that the presence of higher amounts of thyroxine can actually drive the female into anestrus. The lower

concentration of IGF-1 in the moderate conditioned mares is due to the lowered body weight of these mares as IGF-1 plays a role in the maintenance of body weight. The differences in T_4 and IGF-1 concentrations between the 2 groups suggest that if the moderate conditioned mares fell below a BCS 5 that the elevation of thyroxine along with the decline in IGF-1 might be the indicators of potential reproductive inefficiencies. Additionally, the low amounts of body fat of mares in a BCS lower than 5 would possibly drive the serum concentrations of leptin below the levels found in mares of this study. This could also cause lowered reproductive efficiency.

All in all, this study finds that mares expected to breed and conceive shortly after giving birth should be maintained in a BCS of 5 or higher, therefore optimizing time to ovulation and conception rates.

LITERATURE CITED

Adashi, E. Y. 1998. The IGF family and folliculogenesis. J. Reprod. Immun. 39:13-19.

Ahima, R. S., D. Prabakaran, C. Mantzoro, D. Qu, B. Lowell, E. Maratos-Flier, and J.S. Flier. 1996. Role of leptin in the neuroendocrine response to fasting. Nature 382: 250-252.

Allen, W. R., S. Wilsher, C. Turnbull, F. Stewart, J. Ousey, P. D. Rossdale, and A. L. Fowden. 2002. Influence of maternal size on placental, fetal and postnatal growth in the horse. I. Development *in utero*. Reproduction 123:445-453.

Amino, N., T. Yamada, T. Mitsuma, T. Nogimori, O. Tanizawa, M. Kawashima, K. Kurachi, and K. Miyai. 1981. Increase in plasma thyrotropin-releasing hormone in normal human pregnancy. J. Clin. Endoc. Met. 53:1288-1290.

Amstalden, M., M. R. Garcia, S. W. Williams, R. L. Stanko, S. E. Nizielski, C. D. Morrison,D. H. Keisler, and G. L. Williams. 2002. Leptin gene expression, circulating leptin, and luteinizing hormone pulsatility are acutely responsive to short-term fasting in prepubertal heifers: relationships to circulating insulin and insulin-like growth factor I. Biol. Reprod. 63:127-133.

Anthony, R. V., R. A. Bellows, R. E. Short, R. B. Staigmiller, C. C. Kaltenbach, and T. G. Dunn. 1986. Fetal growth of beef calves. I. Effect of prepartum dietary crude protein on birth weight, blood metabolites and steroid hormone concentrations. J. Anim. Sci. 62:1363-1374.

Appleton, D. J., J. S. Rand, and G. D. Sunvold. 2002. Plasma leptin concentrations are independently associated with insulin sensitivity in lean and overweight cats. J. Feline Med. Surg. 4:83-93.

Barash, I. A., C. C. Cheung, D. S. Weigle, H. Ren, E. B. Kabigting, J. L. Kuijper, D. K. Clifton, and R. A. Steiner. 1996. Leptin is a metabolic signal to the reproductive system. Endocrinology 137(7):3144-3147.

Beam, S. W., and W. R. Butler. 1999. Effects of energy balance on follicular development and first ovulation in postpartum dairy cows. J. Reprod. Fertil. Suppl. 54:411-424.

Bidwell, C. A., S. Q. Ji, G. R. Frank, S. G. Cornelius, G. M. Willis, and M. E. Spurlock. 1997. Cloning and expression of the porcine obese gene. Anim. Biotech. 8: 191-206.

Blache, D., R. L. Tellam, L. M. Chagas, M. A. Blackberry, P. E. Vercoe, and G. B. Martin. 2000. Level of nutrition affects leptin concentrations in plasma and Cerebrospinal fluid in sheep. J. Endocrinol. 165:625-637.

Blum, J. W., M. Gingins, W. Schnyder, P. Kunz, E. F. Thomson, P. Vitins, A. Blom, A. Burger, and H. Bickel. 1979. Energy intake in ruminants: effects on blood plasma levels of hormones and metabolites. Inter. J. Vit. Nutr. Res. 49:121-122.

Bray, G. A. 1996. Leptin and leptinomania. The Lancet 348:140-141.

Brown, J. E., H. N. Jakobson, L. H. Askue, and M. G. Peick. 1981. Influence of pregnancy weight gain on the size of infants born to underweight women. Obstet. Gynecol. 57:13-17.

Buff, P. R., A. C. Dodds, C. D. Morrison, N. C. Whitley, E. L. McFadin, J. A. Daniel, J. Djiane, and D. H. Keisler. 2002. Leptin in horses: Tissue localization and relationship between peripheral concentrations of leptin and body condition. J. Anim. Sci. 80:2942-2948.

Buonomo, F. C., and C. A. Baile. 1991. Influence of nutritional deprivation on insulin like growth factor I, somatotropin, and metabolic hormones in swine. J. Anim. Sci. 69:755-760.

Calandra, C., D. A. Abell, and N. A. Belscher. 1981. Maternal obesity in pregnancy. Obstet. Gynec. 57:8-12.

Cammisotto, P. G., and L. J. Bukowiecki. 2002. Mechanisms of leptin secretion from white adipocytes. Am. J. Physiol. Cell Physiol. 283:C244-C250.

Carnevale, E. M., and O. J. Ginther. 1992. Relationships of age to uterine function and reproduction efficiency in mares. Theriogenology 37:1101-1115.

Cartmill, J. A., D. L. Thompson, Jr., W. A. Storer, L. R. Gentry, and N. K. Huff. 2003. Endocrine responses in mares and geldings with high body condition scores grouped by high vs. low resting leptin concentrations. J. Anim. Sci. 81:2311-2321.

Chung, C. S., T. D. Etherton, and J. P. Wiggins. 1985. Stimulation of swine growth by porcine growth hormone. J. Anim. Sci. 60:118-130.

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

Considine, R. V., M. K. Sinha, M. L. Heiman, A. Kriauciunas, T. W. Stephens, M. R. Nyce, J. P. Ohannesian, C. C. Marco, L. J. McKee, T. L. Bauer, and J. F. Caro. 1996. Serum immunoreactive-leptin concentrations in normal-weight and obese humans. N. Engl. J. Med. 334:292-295.

Dahl, G. E., N. P. Evans, L. A. Thrun, and F. J. Karsch. 1995. Thyroxine is permissive to seasonal transitions in reproductive neuroendocrine activity in the ewe. Biol. Reprod. 52:690-696.

Danforth, E., E. S. Horton Jr., M. O'Connell, E. A. H. Sims, A. G. Burger, S. H. Ingbar, L. Braverman, and A. G. Vagenakis. 1979. Dietary-induced alterations in thyroid hormone metabolism during overnutrition. J. Clin. Investigation 64:1336-1347.

DeRouen, S. M., D. E. Franke, D. G. Morrison, W. E. Wyatt, D. F. Coombs, T. W. White, P. E. Humes, and B. B. Greene. 1994. Prepartum body condition and weight influences on reproductive performance of first-calf beef cows. J. Anim. Sci. 72:1119-1125.

Dunn, T. G., J. E. Ingalls, D. R. Zimmerman, and J. N. Wiltbank. 1969. Reproductive performance of 2-year-old Hereford and Angus heifers as influenced by pre- and postcalving energy intake. J. Anim. Sci. 29:719-726.

Dunn, T. G., and C. C. Kaltenbach. 1980. Nutrition and postpartum interval of the ewe, sow and cow. J. Anim. Sci. (Suppl. 2):51:29-38.

Ekbald, U., and S. Grenman. 1992. Maternal weight, weight gain during pregnancy and pregnancy outcome. Int. J. Gynec. Obstet. 39:277-283.

Emilsson, V., Y. Liu, M. A. Cawthorne, N. M. Morton, and M. Davenport. 1997. Expression of the functional leptin receptor mRNA in pancreatic islets and direct inhibitory action of leptin on insulin secretion. Diabetes 46:313-316.

Evans, J. W. 1991. Biorhythms in plasma progesterone concentrations and absence of any correlation to LH biorhythms during different stages of the equine oestrous cycle. J. Reprod. Fertil. (Suppl.):44: 684-685.

Federation of Animal Science Societies. 1999. First Revised Edition. Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching, Savoy, IL.

Fitzgerald, B. P., H. I'Anson, S. J. Legan, and R. G. Loy. 1985. Changes in patterns of luteinizing hormone secretion before and after the first ovulation in the postpartum mare. Biol. Reprod. 33:316-323.

Fitzgerald, B. P., and C. J. McManus. 2000. Photoperiodic versus metabolic signals as determinants of seasonal anestrus in the mare. Biol. Reprod. 63:335-340.

Fitzgerald, B. P., S. E. Ready, D. R. Sessions, M. M. Vick, and B. A. Murphy. 2003. Obesity disrupts the duration of the estrous cycle in the mare. J. Anim. Sci. (Suppl. 81): 102. (Abstr.)

Follett, B. K., and C. Potts. 1990. Hypothyroidism affects reproductive refractoriness and the seasonal oestrus period in Welsh mountain ewes. J. Endoc. 127:103-109.

Folman, Y., M. Rosenberg, Z. Herz, and M. Davidson. 1973. The relationship between plasma progesterone concentrations and conception in post-partum dairy cows maintained on two levels of nutrition. J. Reprod. Fert. 34:267-278.

Frisch, R. E., and J. W. McArthur. 1974. Menstrual cycles: Fatness as a determinant of minimum weight necessary for their maintenance or onset. Science 185:949-951.

Garcia, M. C., and O. J. Ginther. 1978. Regulation of plasma LH by estradiol and progesterone in ovariectomized mares. Biol. Reprod. 19:447-453.

Garcia, M. R., M. Amstalden, S. W. Williams, R.L. Stanko, C. D. Morrison, D. H. Keisler, S. E. Nizielski, and G. L. Williams. 2002. Serum leptin and its adipose gene expression during pubertal development, the estrous cycle, and different seasons in cattle. J. Anim. Sci. 80:2158-2167.

Gentry, L. R., D. L. Thompson, Jr., G. T. Gentry, Jr., K. A. Davis, and R. A. Godke. 2002a. High versus low body condition in mares: Interactions with responses to somatotropin, GnRH analog, and dexamethasone. J. Anim. Sci. 80:3277-3285.

Gentry, L. R., D. L. Thompson, Jr., G. T. Gentry, Jr., K. A. Davis, R. A. Godke, and J. A. Cartmill. 2002b. The relationship between body condition, leptin, and reproductive and hormonal characteristics of mares during the seasonal anovulatory period. J. Anim. Sci. 80:2695-2703.

Gibbs, P. G., and K. E. Davison. 1992. A field study on reproductive efficiency of mares maintained predominately on native pasture. J. Eq. Vet. Sci. 12 (4):219-222.

Ginther, O. J. 1992. Reproductive biology of the mare. Basic and applied aspects. Page 234 in 2nd ed. Equiservices, Cross Plains, WI.

Ginther, O. J., E. L. Gastal, M. O. Gastal, and M. A. Berg. 2004. Critical role of insulin-like growth factor system in follicle selection and dominance in mares. Biol. Reprod. 70:1374-1379.

Glade, M. J., and T. J. Reimers. 1985. Effects of dietary energy supply on serum thryoxine, tri-iodothyronine and insulin concentrations in young horses. J. Endoc. 104:93-98.

Goater, L. H., T. N. Meacham, F. C. Gwazdauskas, and J. P. Fontenot. 1981. The effect of feeding excess energy to mares during late gestation. Page 111 in Proc. 7th Equine Nutr. Phys. Soc., Virginia Polytechnical Institute, Blacksburg.

Hartz, A. J., P. N. Barboriak, A.Wong, K. P. Katayama, and A. A. Rimm. 1979. The association of obesity with infertility and related menstrual abnormalities in women. Int. J. Obesity 3:57-73.

Heidler, B., H. Sauerwein, U. Heintges, J. Aurich, W. Pohl, and C. Aurich. 2002. Metabolic profiles and plasma leptin concentrations in lactating and non-lactating mares. Theriogenology 58:557-561.

Henneke, D. R., G. D. Potter, and J. L. Kreider. 1981. Rebreeding efficiency in mares fed different levels of energy during late gestation. Page 101 in Proc. 7th Equine Nutr. Phys. Soc., Virginia Polytechnical Institute, Blacksburg.

Henneke, D. R., G. D. Potter, J. L. Kreider, and B. F. Yeates. 1983. Relationship between condition score, physical measurements and body fat percentage in mares. Equine Vet. J. 15(4):371-372.

Henneke, D. R., G. D. Potter, and J. L. Kreider. 1984. Body condition during pregnancy and lactation and reproductive efficiency in mares. Theriogenology 21(6):897-909.

Hess, B. W., S. L. Lake, E. J. Scholljegerdes, T. R. Weston, V. Nayigijugu, J. D. C. Molle, and G. E. Moss. 2005. Nutritional controls of beef cow reproduction. J. Anim. Sci. 83 (E. Suppl.):E90-E106.

Hines, K. K. 1985. Reproductive efficiency and endocrine status of postpartum mares at different levels of body condition. Ph.D. Diss., Texas A&M Univ., College Station.

Hines, K. K., S. L. Hodge, J. L. Kreider, G. D. Potter, and P. G. Harms. 1987. Relationship between body condition and levels of serum luteinizing hormone in postpartum mares. Theriogenology 28 (6):815-825.

Houghton, P. L., R. P. Lemenager, L. A. Horstman, K. S. Hendrix, and G. E. Moss. 1990. Effects of body composition, pre- and postpartum energy level and early weaning on reproductive performance of beef cows and preweaning calf gain. J. Anim. Sci. 68:1438-1446.

Houseknecht, K. L., D. L. Boggs, D. R. Campion, J. L. Sartin, T. E. Kiser, G. B. Rampacek, and H. E. Amos. 1988. Effect of dietary energy source and level on serum growth hormone, insulin-like growth factor 1, growth and body composition in beef heifers. J. Anim. Sci. 66:2916-2923.

Houseknecht, K. L., C. A. Baile, R. L. Matter, and M. E. Spurlock. 1998. The biology of leptin: A review. J. Anim. Sci. 76:1405-1420.

Houseknecht, K. L., C. P. Portocarrero, S. Ji, R. Lemenager, and M. E. Spurlock. 2000. Growth hormone regulates leptin gene expression in bovine adipose tissue: Correlation with adipose IGF-1 expression. J. Endo. 164:51-57.

Humphrey, W. D., C. C. Kaltenbach, T. G. Dunn, D. R. Koritnik, and G. D. Niswender. 1983. Characterization of hormonal patterns in the beef cow during postpartum anestrus. J. Anim. Sci. 56 (2):445-452.

Ingram, D. L., and S. E. Evans. 1980. Dependence of thryoxine utilization rate on dietary composition. British J. Nutr. 43:525-531.

Johnson, A. L. 1986. Serum concentrations of prolactin, thyroxine and triiodothryonine relative to season and the estrous cycle in the mare. J. Anim. Sci. 62:1012-1020.

Johnson, C. A., D. L. Thompson, and J. A. Cartmill. 2003. Effects of deslorelin acetate implants in horses: Single implants in stallions and steroid-treated geldings and multiple implants in mares. J. Anim. Sci. 81:1300-1307.

Kane, R. A., M. Fisher, D. Parrett, and L. M. Lawrence. 1987. Estimating fatness in horses. Page 127 in Proc. 10th Equine Nutr. Phys. Symp., Ft. Collins, CO.

Karsch, F. S. 1987. Central action of ovarian steroids in the feedback regulation of pulsatile secretion of luteinizing hormone. Annu. Rev. Physiol. 49:365-382.

Karsch, F. J., G. E. Dahl, T. M. Hachigian, and L. A. Thrun. 1995. Involvement of thyroid hormones in seasonal reproduction. J. Repro. Fert. (Suppl.) 49:409-422.

Kelley, S. T., F. W. Oehme, and G. W. Brandt. 1974. Measurement of thyroid gland function during the estrous cycle of nine mares. Amer. J. Vet. Res. 35:657-660.

Kieffer, T. J., R. S. Heller, C. A. Leech, G. G. Holz, and J. F. Habener. 1997. Leptin suppression of insulin secretion by the activation of ATP-sensitive K+ channels in pancreatic β–cells. Diabetes 46:1087-1093.

Kiess, W., A. Reich, K. Meyer, A. Glasow, J. Deutsher, J. Klammt, Y. Yang, G. Muller, and J. Kratzsch. 1999. A role for leptin in sexual maturation and puberty. Hormone Res. 51:55-63.

Kiyma, Z., B. M. Alexander, E. A. Van Kirk, W. J. Murdoch, D. M. Hallford, and G. E. Moss. 2004. Effects of feed restriction on reproductive and metabolic hormones in ewes. J. Anim. Sci. 82:2548-2557.

Kolaczynski, J. W., J. P. Ohannesian, R. V. Considine, C. C. Marco, and J. F. Caro. 1996. Response of leptin to short-term and prolonged overfeeding in humans. J. Clin. Endocr. Metab. 81:4162-4165.

Kubiak, J. R., J. W. Evans, G. D. Potter, P. G. Harms, and W. L. Jenkins. 1989. Postpartum reproductive performance in the multiparous mare fed to obesity. Theriogenology 32 (1):27-36.

Lake, S. L., E. J. Scholljegerdes, R. L. Atkinson, V. Nayigihugu, S. I. Paisley, D. C. Rule, G. E. Moss, T. J. Robinson, and B. W. Hess. 2005. Body condition score at parturition and postpartum supplemental fat affects on cow and calf performance. J. Anim. Sci. 83:2908-2917.

Lents, C. A., R. P. Wettemann, F. J. White, I. Rubio, N. H. Ciccioli, L. J. Spicer, D. H. Keisler, and M. E. Payton. 2005. Influence of nutrient intake and body fat on concentrations of insulin-like growth factor-1, insulin, thyroxine, and leptin in plasma of gestating beef cows. J. Anim. Sci. 83:586-596.

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

Lucy, M. C. 2000. Regulation of ovarian follicular growth by somatotropin and insulin-like growth factors in cattle. J. Dairy Sci. 83:1635-1647.

Mazerbourg, S., C. A. Bondy, J. Zhou, and P. Monget. 2003. The insulin-growth factor system: A key determinant role in the growth and selection of ovarian follicles? A comparative species study. Reprod. Domest. Anim. 38:247-258.

McArdle, C. A., and A. P. Holtorf. 1989. Oxytocin and progesterone release from bovine corpus luteal cells in culture; effects of insulin-like growth factor I, insulin, and prostaglandins. Endocrinology 124:1278-1286.

McKinnon, A. O., and J. L. Voss. 1993. Equine Reproduction. Pages 57-62 and 570. Lea & Febiger, Philadelphia, London.

McManus, C. J., and B. P. Fitzgerald. 2000. Effects of a single day of feed restriction on changes in serum leptin, gonadotropins, prolactin, and metabolites in aged and young mares. Dom. Anim. Endo. 19:1-13.

Miller, K. F., S. L. Berg, D. C. Sharp, and O. J. Ginther. 1980. Concentrations of circulating gonadotropins during various reproductive states in mares. Biol. Reprod. 22:744-750.

Moenter, S. M., C. J. I. Woodfill, and F. J. Karsch. 1991. Role of the thyroid gland in seasonal reproduction: Thyroidectomy blocks seasonal suppression of reproductive neuroendocrine activity in ewes. Endocrinology 128:1337-1344.

Montague, C. T., I. S. Farooqi, J. P.Whitehead, M. A. Soos, H. Rau, N. J. Wareham, C.P. Sewter, J. E. Digby, S. N. Mohammed, J. A. Hurst, C. H. Cheetham, A. R. Earley, A. H. Barnett, J. B. Prins, and S. O'Rahilly. 1997. Congenital leptin deficiency is associated with severe early-onset obesity in humans. Nature (Lond.) 387:903-908.

Muller, G., J. Ertl, M. Gerl, and G. Preibisch. 1997. Leptin impairs metabolic actions of insulin in isolated rat adipocytes. J. Biol. Chem. 272 (16):10585-10593.

Muoio, D. M., and G. L. Dohm. 2002. Peripheral metabolic actions of leptin. Best Prac. Res. Clin. Endoc. Met. 16 (4):653-666.

Nagatani, S., R. C. Thompson, and D. L. Foster. 1999. Distinctive roles of leptin on the stress and reproductive axes. Page 285 in 81st Annual Meeting of the Endo. Soc., San Diego, CA.

Nagatani, S., Y. Zeng, D. H. Keisler, and D. L. Foster. 2000. Leptin regulates pulsatile luteinizing hormone and growth hormone secretion in sheep. Endocrinology 141 (11): \3965-3975.

Nett, T. M., B. W. Pickett, and E. L. Squires. 1979. Effects of equimate (ICI-81008) on levels of luteinizing hormone, follicle-stimulating hormone and progesterone during the estrous cycle of the mare. J. Anim. Sci. 48:69-75.

Nippoldt, T. B., N. E. Reame, R. P. Kelch, and J. C. Marshall. 1989. The roles of estradiol and progesterone in decreasing luteinizing hormone pulse frequency in the luteal phase of the menstrual cycle. J. Clin. Endocrinology Metab. 69:67-76.

Noden, P. A., W. D. Oxender, and H. D Hafs. 1975. The cycle of oestrus, ovulation and plasma levels of hormones in the mare. J. Reprod. Fertil. (Suppl.) 23:189-192.

NRC. 1989. Nutrient Requirements of Horses. 5th rev. ed. Natl. Acad. Press, Washington, D.C.

O'Callaghan, D., A. Wendling, F. J. Karsch, and J. F. Roche. 1993. The effect of exogenous thryoxine on timing of seasonal reproductive transitions in ewes. Biol. Reprod. 49:311-315.

Oliver, M. H. , J. E. Harding, B. H. Breier, P. C. Evans, and P. D. Gluckman. 1993. Glucose but not a mixed amino acid infusion regulates plasma insulin-like growth factor (IGF)-1 concentrations in fetal sheep. Ped. Res. 34:62-65.

Perry, R. C. 1990. Characterization of follicular development and the influence of dietary energy on ovarian dynamics and reproductive function in postpartum anestrous suckled beef cows. Ph.D. Diss., Kansas State University, Manhattan.

Pineda, M. H., O. J. Ginther, and W. H. McShan. 1972. Regression of corpus luteum in mares treated with an antiserum against an equine pituitary fraction. Am. J. Vet. Res. 33:1767-1773.

Plotka, E. D., C. W. Foley, D. M. Witherspoon, G. C. Schmoller, and D. D. Goetsch. 1975. Periovulatory changes in peripheral plasma progesterone and estrogen concentrations in the mare. Am. J. Vet. Res. 36:1359-1362.

Powell, D. M., L. M. Lawrence, D. F. Parrett, J. DiPietro, L. R. Moser, M. G. Fisher, and K. D. Bump. 1989. Body composition changes in broodmares. Page 91 in Proc. 11th Equine Nutr.Phys. Soc., Oklahoma State Univ., Stillwater.

Prewitt, T. E., A. J. D'Ercole, B. R. Switzer, and J. J. Van Wyk. 1982. Relationship of serum immunoreactive somatomedin-C to dietary protein and energy in growing rats. J. Nutr. 112:144-150.

Rae, D. O., W. E. Kunkle, P. J. Chenoweth, R. S. Sand, and T. Tran. 1993. Relationship of parity and body condition score to pregnancy rates in Florida beef cattle. Theriogenology 39:1143-1152.

Rahe, C. H., R. E. Owens, J. L. Fleeger, H. J. Newton, and P. G. Harms. 1980. Patterns of plasma luteinizing hormone in the cyclic cow: Dependence upon the period of the cycle. Endocrinology 107:498-503.

Randel, R. D. 1990. Nutrition and postpartum rebreeding in cattle. J. Anim. Sci. 68: 853-862.

Reimers, T. J., L. K. Mummery, J. P. McCann, R. G. Cowan, and P. W. Concannon. 1984. Effects of reproductive state on concentrations of thyroxine, 3,5,3'-triiodothyronine and corisol in serum of dogs. Biol. Reprod. 31:148-154.

Rhodes, F. M., K. W. Entwistle, and J. E. Kinder. 1996. Changes in ovarian function and gonadotropin secretion preceding the onset of nutritionally induced anestrus in bos indicus heifers. Biol. Repro. 55:1437-1443.

Richards, M. W., J. C. Spitzer, and M. B. Warner. 1986. Effect of varying levels of postpartum nutrition and body condition at calving on subsequent reproductive performance in beef cattle. J. Anim. Sci. 62:300-306.

Richards, M. W., R. P. Wettemann, and H. M. Schoenemann. 1989. Nutritional anestrus in beef cows: Body weight change, body condition, luteinizing hormone in serum and ovarian activity. J. Anim. Sci. 67:1520-1526.

Rutter, L. M., and R. D. Randel. 1983. Postpartum nutrient intake and body condition: Effect on pituitary function and onset of estrus in beef cattle. J. Anim. Sci. 58: 265-273.

Saad, M. F., M. G. Riad-Gabriel, A. Khan, A. Sharma, R. Michael, S. D. Jinagouda, R. Boyadjian, and G. M. Steil. 1998. Diurnal and ultradian rhythmicity of plasma leptin: Effects of gender and adiposity. J. Clin. Endo. Met. 83:453-459.

Schillo, K. K. 1992. Effects of dietary energy on control of luteinizing hormone secretion in cattle and sheep. J. Anim. Sci. 70:1271-1282.

Schneider, J. E., M. D. Goldman, S. Tang, B. Bean, H. Ji, and M. I. Friedman. 1998. Leptin indirectly affects estrous cycles by increasing metabolic fuel oxidation. Hormones and Behavior 33:217-228.

Schwartz, M. W., D. G. Baskin, T. R. Bukowski, J. L. Kuijper, D. Foster, G. Lasser, D. E. Prunkard, D. Porte, Jr., S.C. Woods, R.J. Seeley, and D.S. Weigle. 1996. Specificity of leptin action on elevated blood glucose levels and hypothalamic nueropeptide Y gene expression in ob/ob mice. Diabetes 45:531-535.

Shi, Z. D., and G. K. Barrell. 1994. Thyroid hormones are required for expression of seasonal changes in reed deer (Cervus elaphus) stags. Reprod. Fert. Dev. 6: 187-192.

Sir-Petermann, T., M. Maliqueo, A. Palomino, D. Vantman, S. E. Recabarren, and L. Wildt. 1999a. Episodic leptin release is independent of luteinizing hormone secretion. Human Reprod. 14:2695-2699.

Sir-Peterman, T. M., V. Piwonka, F. Perez, M. Maliqueo, S. E. Recabarren, and L. Wildt. 1999b. Are circulating leptin and luteinizing hormone synchronized in patients with polycystic ovary syndrome? Human Reprod. 14:1435-1439.

Spicer, L. J., W. B. Tucker, and G. D. Adams. 1990. Insulin-like growth factor-I in dairy cows: Relationships among energy balance, body condition, ovarian activity, and estrous behavior. J. Dairy Sci. 73:929-937.

Spicer, L. J., C. C. Chase, Jr., and L. M. Rutter. 2002. Relationship between serum insulin-like growth factor-I and genotype during the postpartum interval in beef cow. J. Anim. Sci. 80:716-722.

Spitzer, J. C., D. G. Morrison, R. P. Wetteman, and L. C. Faulkner. 1995. Reproductive responses and calf birth and weaning weights as affected by body condition at parturition and postpartum weight gain in primiparous beef cows. J. Anim. Sci. 73:1251-1257.

Sticker, L. S., D. L. Thompson, Jr., J. M. Fernandez, L. D. Bunting, and C. L. DePew. 1995. Dietary protein and(or) energy restriction in mares: Plasma growth hormone, IGF-1, prolactin, cortisol, and thyroid hormone responses to feeding, glucose, and epinephrine. J. Anim. Sci. 73:1424-1432.

Tannenbaum, G. S., H. J. Guyda, and B. I. Posner. 1983. Insulin-like growth factors: a role in growth hormone negative feedback and body weight regulation via brain. Nature. 220:77-79.

Thompson, Jr, D. L., L. Johnson, R. L St. George, and F. Garza, Jr. 1986. Concentrations of prolactin, luteinizing hormone and follicle stimulating hormone in pituitary and serum of horses: Effect of sex, season and reproductive state. J. Anim. Sci. 63:854-860.

Towson, D. H., R. A. Pierson, and O. J. Ginther. 1989. Characterization of plasma progesterone concentrations for two distinct luteal morphologies in mares. Theriogenology 32:197-204.

van Rensburg, S. J., and van Niekerk, C. H. 1968. Ovarian function, follicular oestradiol-17β, and luteal progesterone and 20α-hydroxypregn-4-en-3-one in cycling and pregnant equines. Onderstepoort J. Vet. Res. 35:301-318.

Vandehaar, M. J., B. K. Sharma, and R. L. Fogwell. 1995. Effect of dietary energy restriction on the expression of insulin-like growth factor-I in liver and corpus luteum in heifers. J. Dairy Sci. 78:832-841.

Wang, P. S., T. C. Liu, and G. L. Jackson. 1980. Effects of thyroidectomy and thyroxine therapy on biosynthesis and release of luteinizing hormone by rat anterior pituitary glands in vitro. Biol. Reprod. 23:752-759.

Webster, J. R., S. M. Moenter, C. J. I. Woodfill, and F. J. Karsch. 1991. Role of thyroid gland in seasonal reproduction II. Thyroxine allows a season-specific suppression of gonadotropin secretion in sheep. Endocrinology 129 (1):176 183.

Westervelt, R. G., J. R. Stouffer, H. F. Hintz, and H. F. Schryver. 1976. Estimating fatness in horses and ponies. J. Anim. Sci. 43 (4):781-785.

Whitman, R. W. 1975. Weight change, body condition and beef-cow reproduction. Ph.D. Diss., Colorado State Univ., Fort Collins.

Wiley, J. S., M. K. Petersen, R. P. Ansotegui, and R. A. Bellows. 1991. Production from first-calf beef heifers fed a maintenance or low level of prepartum nutrition and ruminally undegradable or degradable protein postpartum. J. Anim. Sci. 69: 4279-4293.

Williams, G. L., J. Kotwica, W. D. Slanger, D. K. Olson, J. E. Tilton, and L. J. Johnson. 1982. Effects of suckling on pituitary responsiveness to gonadotropin-releasing hormone throughout the early post partum period of beef cows. J. Anim. Sci. 54:594-602.

Williams, G. L., M. Amstalden, M. R. Garcia, R. L. Stanko, S. E. Nizielski, C. D. Morrison, and D. H. Keisler. 2002. Leptin and its role in the central regulation of reproduction in cattle. Dom. Anim. Endo. 23:339-349.

Wiltbank, J. N., W. W. Rowden, J. E. Ingalls, K. E. Gregory, and R. M. Koch. 1962. Effect of energy level on reproductive phenomena of mature Hereford cows. J. Anim. Sci. 21:219-225.

Wu, G., F. W. Bazer, T. A. Cudd, C. J. Meininger, and T. E. Spencer. 2004. Maternal nutrition and fetal development. J. Nutr. 134:2169-2172.

Yu, W. H., M. Kimura, A. Walczewska, S. Karanth, and S. M. Mcann. 1997. Role of leptin in hypothalamic-pituitary function. Proc. Natl. Acad. Sci. USA. 94:1023-1028.

APPENDIX

Appendix Table 1A. ANOVA table for body condition score of mares before parturition.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	47.0323	47.0323	459.21	0.0000
Group	1	47.0323	47.0323	459.21	0.0000
Residual	60	6.1452	0.1024		
Total	61	53.1774	0.8718		

Appendix Table 2A. ANOVA table for rump fat thickness of mares before parturition.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	2.0595	2.0595	19.88	0.0000
Group	1	2.0595	2.0595	19.88	0.0000
Residual	60	6.2155	0.1036		
Total	61	8.2750	0.1357		

Appendix Table 3A. ANOVA table for body fat percentage of mares before parturition.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	45.4947	45.4947	19.88	0.0000
Group	1	45.4947	45.4947	19.88	0.0000
Residual	60	137.3000	2.2883		
Total	61	182.7947	2.9966		

Appendix Table 4A. ANOVA table for body condition score of mares after parturition.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	64.6419	64.6419	526.20	0.0000
Group	1	64.6419	64.6419	526.20	0.0000
Residual	65	7.9850	0.1228		
Total	66	72.6269	1.1004		

Appendix Table 5A. ANOVA table for rump fat thickness of mares after parturition.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	3.1316	3.1316	44.22	0.0000
Group	1	3.1316	3.1316	44.22	0.0000
Residual	65	4.6030	0.0708		
Total	66	7.7346	0.1172		

Appendix Table 6A. ANOVA table for body fat percentage of mares after parturition.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	69.1780	69.1780	44.22	0.0000
Group	1	69.1780	69.1780	44.22	0.0000
Residual	65	101.6780	1.5643		
Total	66	170.8580	2.5888		

Appendix Table 7A. ANOVA table for weight change in mares 1 wk prior to 1 d post foaling.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	1350	1350	0.76	0.3916
Group	1	1350	1350	0.76	0.3916
Residual	22	38883.33	1767.42424		
Total	23	40233.33	1749.27536		

Appendix Table 8A. ANOVA table for change in % body fat in mares 1 wk prior to 1 d post foaling.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	1.325399	1.325399	5.39	0.0299
Group	1	1.325399	1.325399	5.39	0.0299
Residual	22	5.412049	0.246002		
Total	23	6.737449	0.292932		

Appendix Table 9A. ANOVA table for days from parturition to first ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	17.8818	17.8818	1.35	0.2586
Group	1	17.8818	17.8818	1.35	0.2586
Residual	21	278.5530	13.2644		
Total	22	296.4348	13.4743		

Appendix Table 10A. ANOVA table for days from first ovulation to second ovulation postpartum.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	11.6416	11.6416	1.01	0.3259
Group	1	11.6416	11.6416	1.01	0.3259
Residual	21	241.2174	11.5036		
Total	22	253.2174	11.5099		

Appendix Table 11A. ANOVA table for comparison of number of confirmed pregnancies between fat- vs moderate-conditioned mares.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	0.04167	0.04167	0.35	0.5575
Group	1	0.04167	0.04167	0.35	0.5575
Residual	22	2.5833	0.1174		
Total	23	2.6250	0.1141		

Appendix Table 12A. ANOVA table for foal weight measured on d of birth from fat and moderate conditioned mares.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	181.5	181.5	0.69	0.4162
Group	1	181.5	181.5	0.69	0.4162
Residual	22	5814.5	264.295455		
Total	23	5996	260.695652		

Appendix Table 13A. ANOVA table for foal height measured on d of birth from fat and moderate conditioned mares.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	3.375	3.375	1.40	0.2490
Group	1	3.375	3.375	1.40	0.2490
Residual	22	52.95833	2.40719697		
Total	23	56.333333	2.44927536		

Appendix Table 14A. ANOVA table for foal length measured on d of birth from fat and moderate conditioned mares.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	1	0.375	0.375	0.15	0.6985
Group	1	0.375	0.375	0.15	0.6985
Residual	22	53.583333	2.43560606		
Total	23	53.9583333	2.34601449		

Appendix Table 15A. ANOVA table for mean serum concentrations of LH in fat- and moderate-conditioned mares during the interval from foaling to first ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	9	16.699	1.855	1.73	0.0908
Group	1	2.133	2.133	1.99	0.1614
Time	4	13.237	3.309	3.08	0.0190
GroupXTime	4	1.329	0.3323	0.31	0.8711
Residual	110	118.075	1.073		
Total	119	134.775	1.133		

Appendix Table 16A. ANOVA table for mean serum concentrations of LH in fat- and moderate-conditioned mares during the interovulatory interval.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	35	65.298	1.866	3.73	0.0000
Group	1	1.667	1.667	3.33	0.0696
Time	18	58.745	3.264	6.52	0.0000
GroupXTime	16	3.108	0.194	0.39	0.9840
Residual	185	92.556	0.500		
Total	220	157.853	0.718		

Appendix Table 17A. ANOVA table for mean serum concentrations of LH in fat- and moderate-conditioned mares during the interval from foaling to second ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	13	43.068	3.313	7.15	0.0000
Group	1	2.789	2.789	6.02	0.0153
Time	6	37.913	6.319	13.64	0.0000
GroupXTime	6	1.088	0.181	0.39	0.8836
Residual	144	66.721	0.463		
Total	157	109.789	0.699		

Appendix Table 18A. ANOVA table for mean serum concentrations of progesterone in fat- and moderate-conditioned mares during the interval from foaling to first ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	5	342.347	68.469	48.26	0.0000
Group	1	3.625	3.625	2.55	0.1150
Time	2	337.320	168.660	118.87	0.000
GroupXTime	2	3.833	1.917	1.35	0.2664
Residual	63	89.386	1.419		
Total	68	431.733	6.349		

Appendix Table 19A. ANOVA table for mean serum concentrations of progesterone in fat- and moderate-conditioned mares during the interovulatory interval.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	18	1688.382	93.799	10.21	0.0000
Group	1	5.701	5.701	0.62	0.4321
Time	9	1633.176	181.464	19.75	0.0000
GroupXTime	8	30.503	3.813	0.42	0.9105
Residual	148	1359.658	9.187		
Total	166	3048.040	18.362		

Appendix Table 20A. ANOVA table for mean serum concentrations of progesterone in fat- and moderate-conditioned mares during the interval from foaling to second ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	9	812.071	90.230	8.71	0.0000
Group	1	6.627	6.627	0.64	0.4256
Time	4	770.574	192.643	18.59	0.0000
GroupXTime	4	34.871	8.718	0.84	0.5018
Residual	110	1139.657	10.361		
Total	119	1951.728	16.401		

Appendix Table 21A. ANOVA table for mean serum concentrations of leptin in fat- and moderate-conditioned mares during the interval from foaling to first ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	5	1.667	0.333	0.17	0.9744
Group	1	0.2939	0.294	0.15	0.7037
Time	2	0.6744	0.337	0.17	0.8462
GroupXTime	2	0.6978	0.349	0.17	0.8414
Residual	66	132.980	2.015		
Total	71	134.646	1.896		

Appendix Table 22A. ANOVA table for mean serum concentrations of leptin in fat- and moderate-conditioned mares during the interovulatory interval.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	18	10.290	0.572	0.51	0.9492
Group	1	0.743	0.743	0.67	0.4159
Time	9	8.212	0.912	0.82	0.6009
GroupXTime	8	1.797	0.225	0.20	0.9903
Residual	148	165.197	1.116		
Total	166	175.487	1.057		

Appendix Table 23A. ANOVA table for mean serum concentrations of leptin in fat- and moderate-conditioned mares during the interval from foaling to second ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	9	0.492	0.055	0.07	0.9999
Group	1	0.027	0.027	0.04	0.8519
Time	4	0.241	0.060	0.08	0.9888
GroupXTime	4	0.224	0.056	0.07	0.9903
Residual	110	84.847	0.771		
Total	119	85.339	0.717		

Appendix Table 24A. ANOVA table for mean serum concentrations of thyroxin in fat- and moderate-conditioned mares during the interval from foaling to first ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	5	1.533	0.307	1.84	0.1178
Group	1	1.280	1.280	7.66	0.0073
Time	2	0.111	0.055	0.33	0.7188
GroupXTime	2	0.143	0.071	0.43	0.6545
Residual	66	11.022	0.167		
Total	71	12.555	0.177		

Appendix Table 25A. ANOVA table for mean serum concentrations of thyroxin in fat- and moderate-conditioned mares during the interovulatory interval.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	18	5.586	0.310	2.53	0.0012
Group	1	1.574	1.574	12.80	0.0005
Time	9	1.517	0.169	1.37	0.2061
GroupXTime	8	0.364	0.045	0.37	0.9351
Residual	148	18.190	0.123		
Total	166	23.777	0.143		

Appendix Table 26A. ANOVA table for mean serum concentrations of thyroxin in fat- and moderate-conditioned mares during the interval from foaling to second ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	9	4.964	0.552	4.28	0.0001
Group	1	3.300	3.300	25.61	0.0000
Time	4	0.991	0.248	1.92	0.1117
GroupXTime	4	0.673	0.168	1.31	0.2726
Residual	110	14.176	0.129		
Total	119	19.140	0.161		

Appendix Table 27A. ANOVA table for mean serum concentrations of IGF-1 in fat- and moderate-conditioned mares during the interval from foaling to first ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	5	51612.588	10322.518	2.58	0.0343
Group	1	43311.152	43311.152	10.82	0.0016
Time	2	5651.460	2825.730	0.71	0.4974
GroupXTime	2	2649.976	1324.988	0.33	0.7194
Residual	66	264205.035	4003.107		
Total	71	315817.623	4448.136		

Appendix Table 28A. ANOVA table for mean serum concentrations of IGF-1 in fat- and moderate-conditioned mares during the interovulatory interval.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	18	121880.384	6771.132	1.58	0.0726
Group	1	48726.691	48726.691	11.36	0.0010
Time	9	16756.987	1861.887	0.43	0.9150
GroupXTime	8	1608.478	201.060	0.05	1.0000
Residual	148	635091.257	4291.157		
Total	166	756971.641	4560.070		

Appendix Table 29A. ANOVA table for mean serum concentrations of IGF-1 in fat- and moderate-conditioned mares during the interval from foaling to second ovulation.

Source	DF	Partial SS	Mean Square	F Value	Pr>F
Model	9	83691.373	9299.041	1.80	0.0770
Group	1	65796.148	65796.148	12.70	0.0005
Time	4	16152.454	4038.113	0.78	0.5409
GroupXTime	4	1742.771	435.693	0.08	0.9872
Residual	110	569855.605	5180.506		
Total	119	653546.978	5491.991		

Appendix Table 30A. Composition of concentrate used in feeding all mares.

Ingredient Name:

Milo
Wheat midds
Soybean meal-48
Soybean hulls
Liquid binder
Horse premix #6886
Ground lime
Salt mixing

Nutrient	Units	Amount
Weight	LBS	1.00
Protein	%	13.00
Fat	%	2.90
Fiber	%	10.00
Ca	%	0.70
P	%	0.50
ADF	%	13.77
NDF	%	30.69
Lys.	%	0.62
Met.	%	0.18
K	%	0.87
D.M.	%	88.61
S	%	0.17
Mg	%	0.25
Mn	PPM	99.44
Fe	PPM	156.30
Cu	PPM	32.47
Co	PPM	0.79
Zn	PPM	115.13
I	PPM	0.59
Se	PPM	0.44
Vit A	IU/LB	3,019.00
Vit D	IU/LB	210.00
Vit E	IU/LB	38.10
Chol	MG/LB	412.51
Ribo	MG/LB	1.57

Niac	MG/LB	27.94
Pant	MG/LB	6.80
B 12	MCG/LB	3.20
Biot	MCG/LB	149.44
Pyrd	MG/LB	2.50
Thia	MG/LB	5.77
Fo A	MG/LB	0.26
Zn:Cu	PPM	3.55
Dig Lys	PPM	0.34
DE H	KCAL/LB	1,289.62
Ca:P	Ca:P	1.40
Vit A added	IU/LB	2,080.45
Vit D added	IU/LB	27.43
Valine	%	0.42

VITA

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Research Interests: I am most interested in the nutritional effects on reproductive
                         performance in the mare. The hormonal influences of this subject
                         are also of importance. Additionally, I am interested in
                         spermatozoa characteristics of the stallion and the effects of the
                         cooling and storing processes on sperm motility and morphology.