THE EFFECT OF SELECTION FOR MILK YIELD ON NET ENERGY BALANCE AND PLASMA CONCENTRATIONS OF ENDOGENOUS HORMONES AND METABOLITES IN PRIMIPAROUS HOLSTEINS

BY

Alisa Jean Lukes

Thesis submitted to the Graduate Faculty of the Virginia Polytechnic Institute and State University in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

in

Dairy Science

APPROVED:

M.A. Barnes, CHAIRMAN

R.M. Akers

D.M. Denbow

W.E. Vinson
DEPARTMENT HEAD

R.E. Pearson

May, 1988

Blacksburg, Virginia

Endocrine, metabolite and energy balance relationship in Holstein cows of differing genetic merit.

by

Alisa Jean Lukes

Committee Chairman: Michael A. Barnes
Department of Dairy Science

(ABSTRACT)

The effect of genetic selection for milk yield on lactation yield, net energy balance (NEB) and on plasma growth hormone (GH), insulin (INS), prolactin (PRL), nonesterified fatty acids (NEFA) and glucose was studied in primiparous Holstein cows. Net energy balance was calculated and serial blood samples were collected at 0, 45, 90, 180 d postpartum (dpp) and 14 d of the dry period over a 7 h period via jugular cannulae. After 2.5 h of blood collection, growth hormone releasing factor (GRF) was administered at 0, 45, 180 dpp and 14 day of the dry period, while epinephrine was administered at 90 dpp. Nonesterified fatty acids were quantified in 90 dpp samples only. Basal and response periods for each hormone and metabolite were compared by analysis of variance. Milk yield was greater ($P<.05$), NEB was decreased ($P<.05$) and plasma GH was greater ($P<.05$) in selection cows (high-yielders) compared to control cows (low-yielders), while PRL, INS, glucose and NEFA were not different. Growth hormone increased in both groups in response to GRF at all

days postpartum measured, while PRL, INS and glucose were not altered. Epinephrine administration at 90 dpp, stimulated an increase in plasma NEFA, glucose and INS in both groups. Control cows showed a greater ($P<.01$) INS response than selection cows. Results indicate differences exist in GH concentration between genetically selected high- and low-yielding Holsteins during early lactation, but the question remains if these differences are due to energy balance differences or differences in genetic merit for milk yield.

ACKNOWLEDGEMENTS

During my graduate studies in Dairy Science at Virginia Tech, I have had the pleasant opportunity to interact with Dr. Michael A. Barnes as a teacher, advisor, boss and friend. With reassurance, a firm hand and a good joke, Dr. Barnes has instilled in me the desire to set goals and then recognize my ability to achieve them. He has also helped me learn to laugh at myself, and I know that he has had the opportunity to laugh at me too, at least a time or two! I agree with "Kazmer" that one could hardly find a better advisor.

I would also like to thank the members of my committee, Drs. R. M. Akers (not Ackers), D. M. Denbow, R. E. Pearson and W. E. Vinson for being professional, helpful and kind throughout my project. A big THANK-YOU to Chuck Miller, Harold Nester, Bruce Fugate, Dave Page, William (Woody) Saville and others of the farm crew who assisted in my many farm adventures. Also, "Gracias" to Donna Grusell, Dave Whitlock and Beth Grove; all three of these students were excellent help as well as being both humorous and warm.

The times that Willie Foster and Al Pasquino pulled me from the jaws of the computer mumbo-jumbo monster are too numerous to count. These two fellows are not only very intelligent but also invaluable help. Thanks!!

Thanks to Mom and Dad for bringing another red-head into the world. My parents are two very wonderful people, and I'd like to think that I am a composite of their best qualities.

Lastly, I'd like to thank Jonathan Wilson for his wealth of emotional support and excellent scientific advice offered throughout my project. He helped me focus on the "Big Picture" when I got bogged down on the little things. After all, he's my HONEY!!!!

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

Chapter I

INTRODUCTION

There are numerous reports in the literature which have suggested that the determination of hormone or metabolite concentration as indicators of genetic merit for milk yield in dairy cattle may have realistic value as a selection tool. If this tool was effective, it could be used to select for higher yielding cows, therefore saving the producer time and creating more production dollars. However, the major stumbling block to the implementation of this selection technique is identification of what physiological changes occur in genetically superior high-yielding dairy cows as a result of increased selection pressure. Throughout literature, elevated plasma GH concentration has been documented in cows genetically superior for milk yield. However, several researchers hypothesize that this difference is due to the large energy drain that accompanies increased milk yield, instead of genetic selection pressure for milk yield. More research into the question of metabolic versus genetic influence on hormone and metabolite concentrations in dairy cattle is imperative to the future evaluation and acceptance of this physiological selection tool.

To remain competitive with other agricultural enterprises, dairy farmers must continuously work to improve production efficiency. The potential use of a hormone or metabolite concentration as an indicator for genetic merit for milk yield becomes an attractive possibility when one considers evaluating the milk yield potential of animals at a young age. Therefore, this study was designed with the following objectives in order to investigate the effect of genetic selection for milk yield on several physiological and performance traits in primiparous Holsteins:

1. To determine the influence of selection for milk yield on net energy balance (NEB) and endogenous plasma hormone and metabolite concentrations at different stages of lactation and in the dry period.
2. To determine the influence of selection for milk yield on plasma concentrations of growth hormone (GH), insulin (INS), prolactin (PRL), nonesterified fatty acids (NEFA) and glucose in response to a growth hormone releasing factor challenge.
3. To determine the influence of selection for milk yield on the response of NEFA following an epinephrine challenge.

Chapter II

LITERATURE REVIEW

Gorski (1979) suggested that physiological tools have developed to a point where they could also contribute to genetic manipulation, an example being selection of dairy cattle using a physiological factor as an indirect selection criteria for increased milk production. He points out that several universities have established high and low genetic production groups of differing genetic merit with herd phenotypic differences of 20% or more, where the potential for physiological differences being reflected is great and should be taken advantage of. However, Tucker (1981) points out that little information has been gained in this area due to the fact that physiologists and geneticists have made little progress in learning and applying techniques of the other's discipline. Tucker (1981) suggests that researchers in both disciplines must work toward the ultimate goal of controlling genetic variation of a physiological trait in an attempt to enhance production and production efficiency in the dairy cow.

Nutritional influences and their effect on energy balance in the dairy cow should also be considered by the researcher when studying physiological differences in a genetically manipulated herd (Bines and Hart, 1982). Dairy scientists must try to understand how factors affecting

physiological regulation of nutrient partitioning and hormonal release, genetic merit and nutritional status interact with respect to lactation.

The endocrine system plays an essential role in the regulation of mammary development and function (Tucker, 1981; Massri et al., 1985; Sejrsen and Lovendahl, 1986). Therefore, hormones and regulators of hormone secretion are obvious candidates as predictors of genetic merit for milk production and increased production efficiency. The estimation of the heritability of hormone concentration and relationship between the hormone and production traits could lead to the identification and selection of young animals with future milk production potential.

When investigating the possibility that physiological traits may be used as indicators of genetic merit for milk production, Bauman et al. (1985) concluded that first, the physiological factors that contribute to variation in production efficiency must be identified and secondly, the extent that these factors are sensitive to genetic manipulation must be determined. Growth hormone (GH), insulin (INS) and prolactin (PRL) have been investigated as possible candidates which may be altered by genetic selection because of their direct or indirect involvement in the initiation or maintenance of lactation (Hart et al., 1978; Falconer et al., 1980).

The partitioning of nutrients to various body tissues involves two distinct types of regulation: homeostasis, an acute, minute by minute control, and homeorhesis, orchestrated changes in metabolism in the directed partitioning of nutrients to support differing physiological states, for example pregnancy or lactation (Bauman and Currie, 1980). The most pronounced example of homeorhesis in the dairy cow would be where the initiation of lactation dramatically alters the metabolism of many maternal organs to supply the mammary gland with the nutrients necessary for milk synthesis. The nutrient needs of the mammary gland are of such magnitude relative to the total metabolism in high producing cows, that the cow may be considered an appendage on the udder rather than the reverse (Brown, 1969). Therefore, milk secretion proceeds at the expense of other metabolic processes (Hickman, 1980). The commencement and maintenance of a successful lactation are dependent on alterations in metabolism that partition nutrients to the mammary gland. Therefore, regulation of nutrient partitioning by homeorhetic and homeostatic mechanisms is extremely important in insuring a high rate of milk production. Bauman and Currie (1980) contend that the importance of partitioning nutrients to support lactation should not be underestimated, because this physiological state, along with pregnancy, is the

essence of survival of the species as well as being the foundation of the dairy industry.

While digestion, nutrient absorption, maintenance requirement and the utilization of metabolic energy for milk production vary little between animals and remain relatively unaffected by selection for increased milk yield, individual cows vary substantially in the manner in which they partition absorbed nutrients (Swan, 1976; Moe, 1981; Bauman and Currie, 1980; Hart, 1983). Bines and Hart (1978) attribute these variations in nutrient partitioning to be genetic in origin and mediated via differences in the endocrine balance. Based on results from selection studies with goats and sheep, Hickman (1980) reports that one of the best known physiological responses to selection for milk yield is a complex genetic mechanism for maximizing the amount and availability of mobilizable adipose tissue at the time of freshening. During early and mid-lactation, high yielding cows divert dietary energy and mobilize body tissues to meet the high metabolic demands of the lactating udder where as low yielding cows, fed the same ration, partition a varying proportion of nutrients to anabolic processes which increase body weight (Hart et al., 1979).

Studies have related circulating GH concentration to certain metabolic events associated with nutrient partitioning (Bines and Hart, 1978; Bauman et al., 1985),

for example, lipolysis, the mobilization of fat tissue stores in times of energy deficit. Growth hormone has an indirect, metabolic role in lactation through nutrient partitioning, in which GH preserves body protein by inhibiting proteolysis and stimulating incorporation of amino acids into muscle. Growth hormone also stimulates the diversion of glucose and fatty acids away from tissue deposition, thus making them an available energy source (Raben, 1973; Trenkle, 1981). Serum GH concentrations are high in heifers at parturition (Ingalls et al., 1973). Convey (1974) considered this elevation in GH to be associated with changes in the body's metabolism to meet the increased demand for energy and protein at the onset of lactation. Growth hormone remains high until after peak production, approximately 5-8 weeks post partum, and then declines to reach its lowest concentration during the dry period (Convey, 1974).

Hart et al. (1975) reported that plasma GH concentration was high at peak lactation in Holstein cows selected for milk yield and decreased as lactation advanced. However, in low yielding Hereford x Holstein cows, fed the same ration and therefore consuming more energy in relation to their energy output in milk, plasma GH concentration was relatively low throughout lactation. Bauman and Currie (1980) and Bauman et al. (1985) argue

that these endocrine differences were caused by differences in energy balance and therefore possibly are not the physiological basis of the genetic difference. Bauman and Currie (1980) calculated averages from the feed intake and energy utilization data of Moe (1965) who investigated the effect of the level of intake on the utilization of diets by the dairy cow. It has been known for 20 years that peak milk production precedes maximum dietary intake by several weeks and that during the first third of lactation, cows are in a negative energy balance and mobilizing body tissue. These animals did not meet the energy needs of production with dietary intake until approximately 16 weeks postpartum, when milk yield had fallen to less than 80% of peak production. Therefore, (Hart et al., 1975) when the same quantity of ration was fed and the energy intake was similar in both groups, the high yielding cows were being underfed while the low yielding cows were being overfed relative to nutrient requirements. In further studies, Hart (1983) fed superior and inferior producers to similar weight gains in an attempt to equalize nutrient intake relative to requirement, and found no difference in plasma GH concentration. Therefore, he concluded that differences in plasma GH concentration between genetically selected high and low producing cattle were due to differences in energy balance and not due to differences in genetic merit

for milk yield. Kazmer et al. (1986) points out that in Hart's 1983 study, in order to feed both groups to similar weight gains, the low producing cattle's feed intake would have to be restricted, while the high producing cattle would have to be fed ad libitum. This, once again, puts high and low producers in 2 differing and artificially induced physiological states in which varying metabolic influences could exist. Kazmer et al. (1986) reported net energy balance (NEB) was not different between Holstein cows which differed in genetic merit for milk yield. The animals in this experiment were fed ad libitum at all stages of lactation and NEB was calculated at 30, 90 and 200 days postpartum (DPP). Kazmer et al. concluded that observed differences in plasma GH concentrations, greater GH in selection than control cows at 30, 90 and 200 DPP, were not due to differences in NEB between groups. This work therefore suggested that increased plasma GH concentration was one physiological factor responsible for the improved milk yield of offspring of high PD milk sires compared to herdmates from sires with lower genetic transmitting ability for milk production. Others have reported differences in GH concentration of cows with different breeding value within a breed (Bryant and Trigg, 1981; Davey et al., 1983; Flux et al., 1984; Barnes et al., 1985). Bonczek et al. (1986) found elevated GH

concentrations in animals selected for high milk yield during peak, but not during mid-lactation compared to low producers.

Massri et al. (1985) took a different approach in attempting to relate plasma hormone concentration to performance traits such as lactation yield. Instead of looking solely at circulating pituitary hormones, which are subject to fluctuations in concentration, he investigated the pituitary response to hypothalamic releasing hormones. In support of this hypothesis, Land et al. (1981) reported that the magnitude of the pituitary release of lutenizing hormone (LH) following a lutenizing hormone releasing hormone (LHRH) injection was repeatable and also hereitable. The galactopoietic activity of GH was reported by Cotes et al. (1949) nearly four decades ago, and plasma GH concentration is positively associated with milk yields (Hart et al., 1979; Kazmer et al., 1986). Therefore, Massri et al. (1985) investigated the magnitude of the pituitary release of GH following a growth hormone releasing factor (GRF) injection in calves of differing genetic merit for milk yield fed a high energy diet. Massri et al. challenged their treatment groups with a single injection of .1 ug/kg human pancreatic GRF (hpGRF 1-40)NH_2. Human pancreatic GRF(1-44) and its fragments hpGRf(1-24)NH_2, hpGRF(1-29)NH_2, and hpGRF (1-40)NH_2 have

all been documented to cause elevated circulating concentration of GH in the ruminant (Baile et al., 1983; Plouzek et al., 1983; McCutcheon et al. 1984; Mosely et al., 1984; Massri, 1985; Enright et al., 1986; Baile and Buonomo, 1987). Massri et al. (1985) found that the magnitude of the hpGRF(1-40)NH_2 induced GH release did not reflect differences due to genetic potential for milk yield. Both selection and control calves responded with a peak GH concentration of similar magnitude, 10-20 minutes post-injection. Massri et al. noted the important role of GH in fatty acid mobilization from adipose tissue in an energy deficient state, and suggested that the calves' high plane of nutrition could have masked any differences in pituitary responsiveness to GH release. He hypothesized that perhaps dietary energy restriction prior to hpGRF(1-40)NH_2 administration would create a metabolic state in which adipose mobilization predominates, providing a nutritional background more conducive for the expression of any differences in the GH response between selection and control calves. He did find however, that selection calves had greater basal GH concentration than control calves.

Sejrsen and Lovendahl (1986) suggested it was unrealistic to believe that one hormone or metabolite alone can explain a major part of the genetic variation in milk yield potential. They suggest a physiological index of

several hormones and metabolites as a more logical and possibly more reliable criteria for differentiating cattle on the basis of genetic merit for milk yield. Since lactation is regulated by a milieu of hormones, some with opposing actions (Tucker, 1981; Bines and Hart, 1982), measurement of INS, PRL, glucose or nonesterified fatty acids (NEFA) should be considered in a genetic merit index.

The role of INS in control of nutrient utilization is essentially anabolic. Insulin stimulates glucose uptake and utilization by many peripheral tissues while inhibiting glucose synthesis and release from the liver (Bines and Hart, 1982). Synthesis of protein and lipid are both stimulated by INS, while proteolysis and lipolysis are inhibited (Basset, 1975). Thus, this protein hormone is associated with many processes that divert energy away from milk synthesis and toward body tissue (Yang and Baldwin, 1973). Changes in plasma INS concentration in dairy cattle are positively correlated with changes in body weight. Also, treatment of lactating cows with INS causes an immediate decrease in milk yield which can be reversed by infusing glucose (Kronfeld, 1963). The effect of changes in the GH:INS ratio on fat mobilization has also been investigated (Rabinowitz et al., 1966). Rabinowitz et al. (1966) proposed that in the human, human GH (hGH) potentially has both an anabolic role, for example,

promotion of protein synthesis, and a diabetigenic role, for example, promotion of hyperglycemia and fat mobilization. He hypothesized that the anabolic actions of hGH are maximized by the presence of INS, conversely, in the absence of INS or in the presence of very low concentrations of INS, the diabetigenic actions of hGH are paramount. Further investigation into the role of the INS:GH ratio in ruminants is needed.

Insulin has been proposed as having an opposing role to GH in nutrient partitioning (Bines and Hart, 1982). In contrast to blood GH concentration in dairy cattle, plasma INS concentration is low in early lactation and increases as lactation progresses (Swan, 1976; Hart et al., 1978). Vasilatos and Wangsness (1981) concluded that high GH and low INS plasma concentration were important factors in maximizing milk production in early lactation in dairy cattle. Comparison of circulating levels of INS in high and low yielding cattle (Hart et al., 1975; Hart et al., 1978; Hart et al., 1979) revealed that during early lactation, INS was significantly greater in the plasma of low yielding cows, which were in a state of energy surplus and gaining weight, compared to high yielding cows, which were in a state of energy deficit and losing weight. Insulin concentration became comparable between the two groups only when lactation ended, indicating a possible

influence of energy balance on INS concentration in cows of differing genetic potential for milk yield. Hart et al. (Hart et al., 1980) reported that INS differences between groups of cows differing in genetic merit for milk yield were due to differences in the INS secretion rate. In more recent studies, Barnes et al. (1985) found INS concentration to be similar between Holstein cattle of differing genetic merit for milk yield. Bonczek et al. (1986) found INS concentration to be lower during the entire lactation in animals selected for milk yield. Conflicting information exists in literature regarding the relationship between INS concentration and genetic merit for milk yield, and the influence of energy balance on this relationship.

PRL is essential for full lactogenesis in cattle (Akers et al., 1981). However, PRL exerts no galactopoietic effect in cows (Folley and Young, 1940), and once lactation is established in ruminants, the concentration of PRL in the circulation can be greatly decreased without markedly affecting milk secretion (Hart et al., 1973). Prolactin has also been implicated as playing a role in the metabolic regulation of ruminants (Bauman et al., 1979; Forbes et al., 1975; Hart et al., 1978; Tucker, 1981). Forbes et al. (1975) suggested an anabolic role for PRL, because lambs with increased growth rates had greater plasma PRL

concentration. Prolactin either directly or indirectly alters the capacity of muscle for net protein accretion and may decrease lipoprotein lipase activity in adipose tissue (Anguis et al., 1979; Botts et al., 1979; Moe, 1981; Williams et al., 1966).

Kazmer et al. (1986) measured PRL concentration before and after thyrotropin releasing hormone (TRH) administration in two groups of first lactation Holstein cattle differing in genetic merit for milk yield. Kazmer et al. found plasma PRL concentration, before and after TRH administration at several stages of lactation, was unaffected by genetic merit for milk yield. This was in agreement with a report by Bonczek et al. (1986) where PRL was measured at peak and mid-lactation in two groups of Holstein cattle, a high yielding group of daughters of cows mated only to commercially available sires and a low yielding group of daughters of cows mated solely with sires that were breed average for milk yield. Bonczek et al. (1986) reported similar PRL concentrations between high and low yielding Holstein cattle at peak and mid lactation. The lack of a positive relationship between PRL concentration during lactation and genetic selection for milk yield is not surprising, because other investigators have also unsuccessfully correlated basal PRL with increased milk production (Hart et al., 1979; Hart et al.,

1978). Hart et al. (1978) reported the lack of a significant correlation between PRL and genetic merit for milk yield when he measured PRL concentration in 8 high yielding Holstein cows and 8 low yielding Hereford x Sussex cows at 40, 80, 120, and 180 days postpartum (dpp). Hart et al. (1979) conducted a similar study and measured PRL concentration during the dry period in cows of unequal genetic merit for milk production. Hart et al. (1979) concluded that if PRL plays a part in the partitioning of nutrients between body tissue and milk, it is not reflected by differences in the plasma concentration of PRL in high and low yielding dairy cattle, during lactation or the dry period.

Lipogenesis remains low and lipolysis high during early lactation in dairy cattle which are in a state of negative energy balance (Swann, 1976; Collier et al., 1984). When the energy requirement decreases in later lactation, adipose tissue metabolic pathways shift to promote increased storage of lipids (Bauman and Currie, 1980). The synthetic pathways for triglyceride storage are de novo lipogenesis and fatty acid esterification, whereas release is through triglyceride hydrolysis (Collier et al., 1984). These pathways are reciprocals in that lipogenesis predominates during a state of positive energy balance and lipolysis predominates during a state of negative energy

balance. This reciprocity is an example of homeorhetic regulation (Collier et al., 1984). Negative net energy balance and high plasma GH concentrations have been associated with high NEFA concentration in dairy cattle (Hart et al., 1978). Nonesterified fatty acids are used largely by the mammary gland for lipogenesis and as an energy source (Kronfeld, 1976).

Cows in negative energy balance increase mobilization of adipose tissue in response to exogenous bovine GH (bGH) treatment (Peel et al., 1982; Peel et al., 1981). Highly purified preparations of pituitary or recombinantly derived bovine somatotropin, while not causing an acute increase in NEFA plasma concentration, do alter lipid metabolism. The specific adaptations of lipid metabolism depend on the energy balance of the treated cow (Peel and Bauman, 1987). When the milk yield response to the GH treatment caused cows to be in a state of negative energy balance, milkfat content increased so that the response in milkfat yield markedly exceeded the response in milk yield, and in this situation, NEFA plasma concentration was chronically elevated (Eppard et al., 1985; McCutcheon and Bauman, 1986; Peel et al., 1981). When animals were in a state of positive energy balance, milkfat percent was not altered, so the increase in milkfat yield was similar to the increase in milk yield (Peel et al., 1982; Fronk et al.,

1983; Bauman et al., 1985; Eppard et al., 1985;) and blood concentration of NEFA was unchanged (Peel et al., 1982; Peel et al., 1983; Eppard et al., 1985;).

Hart et al. (1978) reported underfed, high yielding cows had greater NEFA than lower yielding, overfed cows, with the most significant difference occurring in early lactation. Low yielding cows showed no difference in NEFA concentration between early lactation and the dry period. Therefore, NEFA plasma concentration seems to be influenced by the energy balance of the animal. McNamara and Hillers (1986) studied lipolytic adaptations of bovine adipose tissue during late pregnancy, lactation and the dry period in Holsteins of differing genetic merit and concluded differently as to the influence of NEB on lipolysis rates. McNamara and Hillers found that feeding a low energy diet for the first 140 d of lactation did not affect adipose tissue lipolytic rates measured in vitro. The dietary restriction resulted in a large decrease in NEB but did not stimulate lipolysis in early lactation. It was concluded that the lipolysis rate was near the physiological maximum for those animals, and that the limit was set according to each individual cow's genetic makeup. These researchers therefore suggested a genetic component in adrenergic regulation of lipolysis in adipose tissue, independent of energy balance, in supporting lactation.

Eiseman et al. (1984) conducted a study to evaluate the administration of bGH to growing animals and to further elucidate the regulatory principles concerning accretion of lipid stores. Six Hereford heifers were fed a pelleted concentrate diet (17.6% crude protein) at 4 h intervals throughout the day. This diet was fed at slightly above the maintenance energy requirement of these animals (0.130 Mcal ME/kg BW , NRC). Daily injections of bGH were given for 14 d, and all measurements were taken on day 11 of treatment. An increase in plasma NEFA concentration and an increase in the irreversible loss of NEFA was seen in the treated heifers. This resembles a similar response seen in lactating cows in negative tissue energy balance when injected with bovine GH (Peel et al., 1982). Eiseman (1984) proposed that this increase in NEFA oxidation may be associated with sparing oxidation of other metabolites, such as amino acids, thus making them available for other productive functions.

Bauman and Currie (1980) proposed that homeorhetic regulators such as bGH influence nutrient partitioning by altering responsiveness of key tissues, such as adipose tissue, to homeostatic signals. McCutcheon and Bauman (1986) examined this hypothesis by treating late lactation Holstein cows with bGH and then measuring the effects of this treatment on the response to an intravenous

epinephrine challenge. A total mixed ration calculated to provide 120% of requirements was fed. The epinephrine challenge was given one day following a 12 d bGH treatment period. Plasma GH concentration had generally returned to baseline concentration when the challenge was administered. Baseline concentrations of NEFA were unaffected by bGH administration, this is consistent with several studies involving bGH administration to cows in a positive energy balance (Peel et al., 1982; Peel et al., 1981). Circulating plasma NEFA and plasma glucose concentrations were elevated by the epinephrine challenge in the cows treated with bGH compared to the non-bGH treated cows. Peak plasma NEFA and glucose concentration were reached 15 min post challenge. McCutcheon and Bauman propose that the increased plasma glucose concentration may result from increased glucose production by the liver through increased rates of gluconeogenesis and glycogenolysis, or a decrease of glucose oxidation by body tissues. The increase in plasma NEFA concentration in response to the epinephrine challenge in the bGH treated cows was not an acute effect requiring the presence of high circulating GH because challenges were made when GH concentration had generally returned to baseline, nor was it caused by treatment induced differences in the energy status of the cows, as NEB remained positive and was not affected by bGH

treatment. McCutcheon and Bauman concluded that the administration of exogenous GH must alter the responsiveness of adipose tissue to an acute lipolytic stimulus, while, at the same time, preserving homeostatic control of circulating concentration of metabolites. This hypothesis agrees with GH's proposed chronic, homeorhetic control of metabolism (Bauman and Currie, 1980).

The availability of glucose for lactose synthesis is one of the primary factors limiting milk production in high yielding cows (Kronfeld et al., 1976). In the ruminant, gluconeogenesis provides up to 90% of the necessary glucose (Young, 1977). Glucose utilization in mammary gland biosynthesis is substantial, as it provides 85% of the carbons in lactose, 70% of glycerol and contributes much of the reducing equivalents in the form of nicotinamide adenine dinucleotide phosphate (NADPH) for lipogenesis (Kronfeld, 1976). The amount of glucose available to the mammary gland is determined by the total amount of glucose available to the animal; essentially the limit set by gluconeogenesis, less the amount used by the nervous system and by peripheral tissues other than the mammary gland (Bines and Hart, 1982). Glucose entry rates increase during early lactation even when intake is held constant (Bennink et al., 1972). This suggests an increase in

glucose production independent of feed intake, which must involve an increased rate of gluconeogenesis.

Several researchers have attempted to correlate plasma glucose concentration with genetic merit for milk yield. Hart et al. (1978) measured glucose concentration in high and low yielding Hereford x Holstein cows at 40, 80, 120 and 180 DPP and also after lactation was finished. Hart failed to demonstrate decreased plasma glucose concentrations throughout lactation in the high yielding Holstein cattle and found that the glucose concentration was 18% greater in both groups when the animals were dry compared with values obtained during early lactation. Barnes et al. (1985) reported that while plasma glucose concentration was different between 6, 12, 18 and 24-month-old Holstein cattle, glucose concentration was similar between genetic selection groups; daughters of cows bred to AI-selected sires had similar concentrations compared to daughters of cows bred randomly to non-AI, nonselected sires within each age group. Flux et al. (1984) investigated the plasma glucose concentration in two groups of New Zeland Friesian cows, differing in genetic merit for fat production. The groups were further divided into two groups and either fed ad libitum or fed a restricted amount equal to 70% of a predetermined ad libitum intake. Glucose concentration was different between the genetic

selection groups only when the 2 groups were underfed. Plasma glucose concentration was significantly decreased in the low fat producing group whose feed intake was restricted.

Availability of blood glucose to the mammary gland may be increased as plasma GH concentration increases, because injections of GH increased lactose in milk without affecting irreversible loss of glucose (Bines and Hart, 1982). Therefore, despite the lack of differences in peripheral glucose between genetically superior and inferior dairy cattle in several studies, partitioning of glucose may vary with respect to genetic merit for milk production (Bines and Hart, 1982), because the partitioning of glucose may be partially regulated by plasma GH concentration.

Chapter III

MATERIALS AND METHODS

Twenty-eight primiparous Holstein cows were housed in an open-sided free stall barn at the Virginia Tech Dairy Center and milked twice daily in a milk parlor. Animals were either daughters of cows bred to selected, commercially available artificial insemination (AI) sires (selection group, n = 15) (weighted mean sire Predicted Difference (PD_{82}) = +420 kg) or second to fifth generation daughters of cows continuously randomly bred to nonselected sires originating in the Virginia Tech dairy herd (control group, n = 13) (Mean estimated sire Predicted Difference (PD_{82}) = -368 kg). Therefore the estimated genetic difference in transmitting ability for the 2 groups of bulls used to sire these cows was 759 kg milk. Daily milk yield for both morning (a.m.) and afternoon (p.m.) milkings was monitored and recorded for each cow. Phenotypic performance during the first lactation of these cattle resulted in a difference in 4% FCM of 632 kg milk, with selection cows producing 6,450 kg milk, and control cows producing 5,818 kg milk when records were corrected to a 4% fat corrected milk (FCM) basis.

All lactating animals were fed the same mixed complete diet (blended corn silage, haylage and concentrate) ad

libitum during all stages of lactation. During the dry period, a lower energy ration (ground orchard grass hay and alfalfa haylage) was fed ad libitum. The lactating ration averaged 49.0% dry matter (DM), 22.5% acid detergent fiber (ADF), 14.8% crude protein (CP), 10.2% digestible protein(DP), 72.6% Total digestible nutrients (TDN) and 1.6 Mcal NE /kg on a dry matter basis. The dry period ration averaged 58.0% DM, 16.2% CP, 11.5% DP, 36.9% ADF, 61.6% TDN and 1.3 Mcal NE/kg DM. Body weight (BW) was recorded weekly. Body weight was converted to metabolic body weight ($MBW = BW^{.75}$) for further use in graphing and energy balance calculations. Data were collected throughout the first lactation, and 3 cows were culled before completion of the study due to reproductive problems. The experiment was begun May, 1986 and completed February, 1988.

Serial blood samples were collected at 0 d, 45 d, 180 d postpartum (dpp), and on the 14 d of the dry period via jugular cannulae inserted 4.5 h prior to sampling. On the day of parturition (0 d), serial blood sampling began 12 h postpartum. Samples were collected into tubes containing 200 ul of a disodium ethylenediamine tetraacetate (Na_2 EDTA, Fisher O-2793) and benzamidine hydrochloride (Fisher 1183094) mixture (234 mg benzamidine hydrochloride and 60 mg Na_2 EDTA per ml saline) at 15 min intervals from 1030 to

1600 h and then half-hourly until 1730 h. At 45, 90, and 180 dpp, lactating cows were milked at 0400 h and 1600 h on the sampling day. At 0 d collection period, 79% of the cows were milked before blood sampling was initiated. Growth hormone releasing factor (GRF 1-44 NH_2 Sigma G-0138) was administered (.2 ug/kg body weight) via jugular cannulae to 0, 45, 180 dpp and 14 d dry period cows at 1300 h. Epinephrine (.7 ug/kg body weight) was administered via jugular cannulae to 90 dpp cows at 1300 h. The basal period represents the first 2.5 h of the blood collection regime and the response period represents the 4.5 h of blood collection after the hormonal challenge was given. Blood was immediately chilled on ice and then centrifuged and plasma stored at -20 C until assayed for hormone and metabolite content.

Feed intake data were recorded from 5 d pre to 2 d post cannulation via an electronic intake monitoring device (Pinpointer, Inc.) at -14, 0, 45, 90, 180 dpp and on the 14 d of the dry period. In the lactating cows, milk yield during the 7 d feed intake data collection period was recorded at each milking, except at parturition when milk yield was recorded from 3 dpp to 8 dpp. Net energy balance was calculated by the method of Custodio et al. (1983) at 0, 45, 90, 180 dpp and on the 14 d of the dry period. To calculate net energy balance (NEB), dry matter feed intake

(DMI), BW and milk yield were used in conjunction with the National Research Council (NRC, 1978) factors concerning energy requirements for maintenance and milk production. The appropriate factor from NRC tables was multiplied by metabolic BW ($BW^{.75}$) during each sampling period (at each postpartum period) to yield the energy requirement for maintenance (energy requirement for maintenance = .08 Mcal NE/kg*$BW^{.75}$). The requirement was increased by 20% as recommended for first lactation animals. Energy requirement for milk yield was calculated by multiplying the mean daily 4% FCM yield during the sampling period by the NRC factor for 4% FCM (energy requirement for milk yield = 4% kg FCM * .74 Mcal NE/kg). Milk fat test data were obtained from Dairy Herd Improvement Association (DHIA) records, using the data from the test day occurring in the same month as the sampling period. Total energy requirement was then calculated as the sum of the requirement for maintenance and the requirement for milk yield. Energy intake was calculated by multiplying the energy content (Mcal/kg DM) of the ration by the daily amount consumed. Finally, NEB was calculated as the difference between energy intake and the total energy requirement.

In dry cows, the procedure for calculation of NEB was different in that the energy requirement for maintenance

during the last 2 months of gestation is greater than during lactation (energy requirement for maintenance during the last 2 months gestation = .104 Mcal NE/kg*$BW^{.75}$). As during lactation, the energy requirement for maintenance was increased by 20% to allow for growth. Net energy balance was calculated as the difference between the energy intake and the energy requirement for maintenance during the last 2 months of gestation.

Plasma PRL concentration was quantified using a double antibody radioimmunoassay (RIA) with bovine PRL antiserum, by the method of Barnes et al. (1985). The specific antibody used at an initial dilution of 1:1500, bound approximately 56% of labelled PRL in the absence of unlabelled hormone. With bovine PRL (NIH-bPRL-6) as a reference standard, the assay system did not cross-react (less than 2%) with GH or INS. Plasma sample volume of 50 or 100 ul were used for blood collected during the summer or winter months, respectively. All samples were assayed in duplicate and the intra- and inter-assay coefficients of variation averaged 6.3% and 7.5% respectively, for two plasma pools. The standard curve ranged from .05 to 5.0 ng and standards added to plasma pools were 84% recoverable.

Plasma GH concentration was quantified using a double antibody RIA with bovine GH antiserum, by the method of Barnes et al. (1985). The specific antibody used at an

initial dilution of 1:500, bound approximately 51% of labelled GH in the absence of unlabelled hormone. With NIH-bGH-18 as a reference standard, the assay system did not cross-react (< 2%) with INS or PRL. All samples, assay volume of 300 ul plasma, were assayed in duplicate and the intra- and inter-assay coefficients of variation averaged 7.8% and 10.5% respectively, in the two plasma pools. The standard curve ranged from .25 to 20 ng and standards added to plasma pools were 84% recoverable

Plasma INS concentration was quantified using a double antibody RIA with bovine INS antiserum, by the method of Barnes et al. (1985). The specific antibody used at an initial dilution of 1:8000, bound approximately 42% of labelled INS in the absence of unlabelled hormone. With Eli Lilly-bINS as a reference standard, the assay system did not cross-react (<2%) with GH or PRL. All samples, assay volume of 300 ul plasma, were assayed in duplicate and the intra- and inter-assay coefficients of variation averaged 10.3% and 10.5% respectively, in the two plasma pools. The standard curve ranged from .05 to 4.0 ng and standards added to plasma pools were 96% recoverable.

Plasma glucose was determined spectrophotometrically (Milton Roy Spectronic 21) using the O-toluidine method of Feteris (1965). The 26 plasma samples collected during a complete collection period from each cow were pooled to

form 7 individual pools. Each pool was composed of a 300 ul aliquot from each sample collected during each consecutive hourly period. One hundred ul of each pool was assayed in duplicate. The standards ranged from 25 mg/dl to 200 mg/dl. The intra- and inter-assay coefficients of variation averaged 5.2% and 8.0% respectively, for one plasma pool.

Plasma nonesterified fatty acid (NEFA) concentration was determined in plasma samples collected 90 dpp, pre- and post- epinephrine challenge, by the acyl CoA synthetase-oxidase method described in the Wako manual (1986) using a split beam spectrophotometer, Bausch and Lomb 1001. The assay system was modified by McCutcheon and Bauman (1986) to allow for reduction in the sample volume (from 50 to 25 ul) and of volume of reagents (to 15% recommended volume). These changes substantially reduced assay costs but did not influence accuracy. The modified system was validated against the methods of Ko and Royer (1967) and Dalton and Kowalski (1967). Plasma from the first 24 blood samples collected from each cow during the 90 dpp collection period were pooled to form 8 individual pools. Pools 1 and 2 were composed of 100 ul aliquots from each of the 4 samples taken during the first and second hr, respectively, of blood sampling. These two pools thus represented the basal period pre-epinephrine challenge NEFA concentration in the

cows. Pools 3, 4, 5, and 6 each represented 4 individual blood samples taken at 15 min intervals immediately following the epinephrine challenge for a total of 1 hr. Pool 7 consisted of 4-100 ul plasma aliquots from blood samples obtained during the second hour post-epinephrine challenge, while pool 8 consisted of 3-100 ul plasma aliquots from blood samples obtained during the third hour post-epinephrine challenge. Blood samples collected from the animals after milking were not included in this assay. Standards of oleic acid in concentrations of 0, 125, 250, 500, and 1000 uEq/l were used. To minimize variation due to low concentration of plasma NEFA, 250 uEq/l of standard solution was added to each sample. Absorbance was recorded at 550 nm and converted to concentration by linear regression of the standard curve. Nonesterified fatty acid concentration of samples was calculated by subtracting 250 uEq/l from each value obtained. The assay was linear to 2000 uEq/l and intra- and inter-assay coefficient of variation were 4% and 1%, respectively.

Data Analysis: Hormonal data were analyzed using the General Linear Models (GLM) option of the Statistical Analysis System (SAS, 1982). Blood hormones and metabolites were analyzed using a model accounting for genetic selection group, cow(genetic selection group), days

postpartum, days postpartum by genetic selection group interaction, period (representing pre- and post-challenge), period by genetic selection group interaction, period by days postpartum interaction, period by days postpartum by genetic selection group interaction, sample(period), genetic selection group by sample(period) interaction and error. Additionally, the mean ambient temperature during the basal or response period was included in the analysis as a covariate. The mean square for cow within genetic selection group was used to test for differences between selection groups. The mean square for error was used to test all other effects. Metabolic body weight, DMI, NEB, and ratio of DMI/FCM were analyzed using a reduced model.

Chapter IV

RESULTS

Selection cows had a greater (P<.05) FCM yield than control cows. Selection cows yielded 6,450 kg FCM and control cows yielded 5,818 kg FCM. Selection cows averaged 303 days in milk (DIM) and control cows averaged 305 DIM. Fat and protein percent were similar between genetic groups, with an average fat test of 3.6% and 3.7% in selection and control cows, respectively, and an average of 3.1% protein in both groups.

Dry matter intake (DMI) increased (P<.01) from -14 d to 90 dpp and was similar at 180 dpp (Figure 1). Dry matter intake was greatly reduced during the dry period compared to lactation (P<.01). There were no significant differences in DMI between the selection and control group cows (Table 1). Dry matter intake of both groups increased by 52% from -14 d to 90 dpp. Intake remained similar between selection and control cows at 180 dpp and then declined 54% in both groups by the 14 d of the dry period (Figure 1).

The ratio of DMI/MBW, indicated that selection group cows were consuming 9.0% of their MBW in DM while control group cows were consuming 11.0% of their MBW in DM at parturition. The ratio increased and peaked at 90 dpp with selection and control cows consuming 16.0% of their

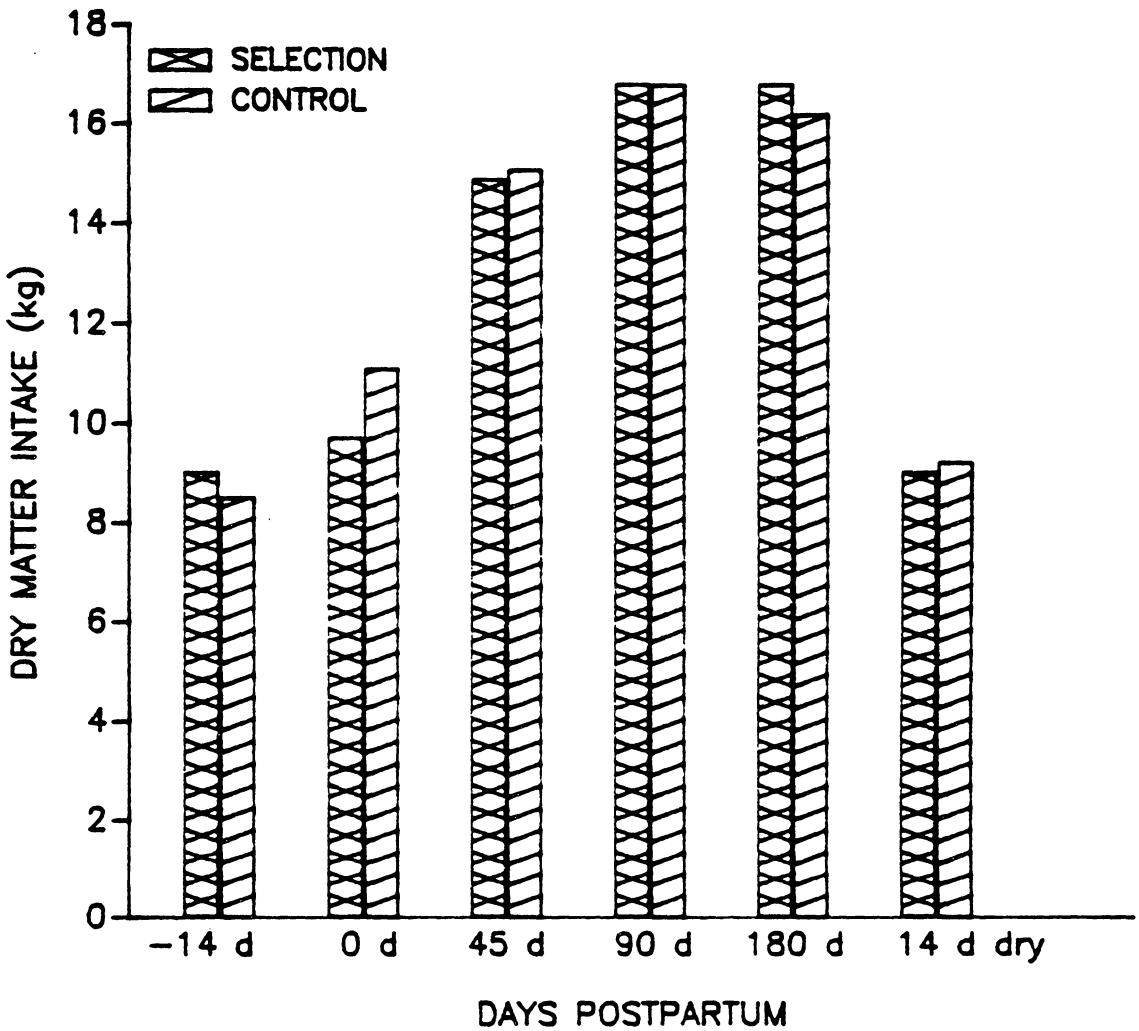

Figure 1. Dry matter intake of selection and control Holstein cows at −14 d, 0, 45, 90, 180 days postpartum and 14 d of the dry period, mean standard error is .4 kg

TABLE 1. Least square means of dry matter intake (DMI), metabolic body weight (MBW), and fat corrected milk (FCM) yield in selection and control cows

Genetic group	Days [1] Postpartum	DMI [2]	MBM [3,4]	FCM [2,5]
selection	0	9.7±.5	107.4±.8	18.5±.6
	45	14.9±.4	102.9±.8	23.1±.6
	90	16.8±.4	106.2±.8	22.0±.6
	180	16.8±.4	111.5±.8	21.0±.6
	dry [6]	9.0±.5	118.5±.8	---
control	0	11.1±.5	104.1±.9	17.3±.7
	45	15.1±.5	103.5±.9	22.0±.7
	90	16.8±.5	107.8±.9	21.0±.7
	180	16.2±.5	113.1±.9	18.2±.7
	dry [6]	9.2±.5	121.3±1.0	---

[1] Days postpartum ($P<.01$) for MBW, DMI, and FCM

[2] Least square means ± SE, kg, 5 d collection mean

[3] Least square means ± SE, MBW = (kg body weight)$^{.75}$

[4] Genetic group * Days postpartum interaction ($P<.01$)

[5] Genetic group ($P<.25$)

[6] Day 14 of the dry period

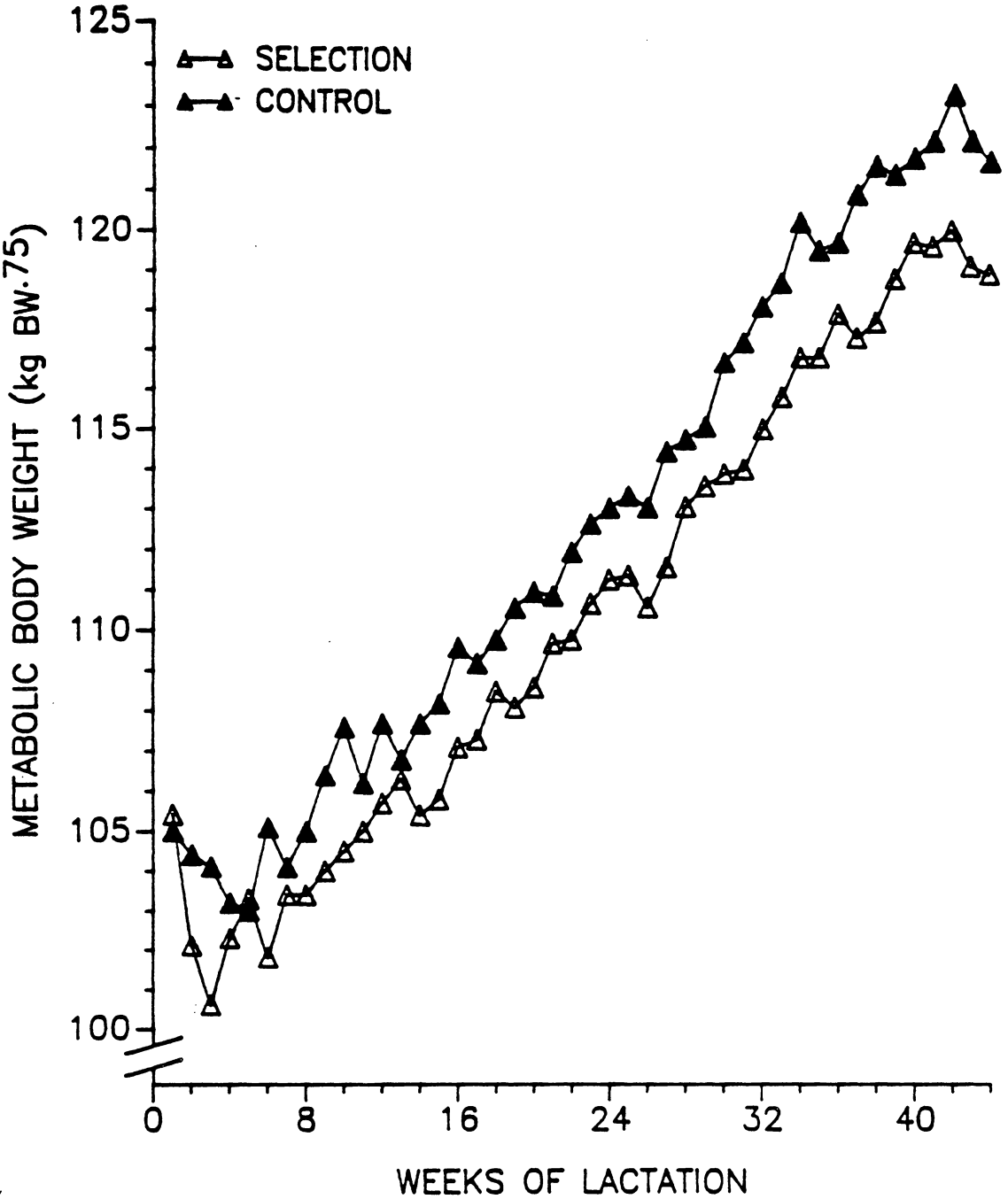

Figure 2. Weekly metabolic body weight (kg body weight$^{\cdot 75}$) of lactating selection and control Holstein cows, mean standard error is .8 kg

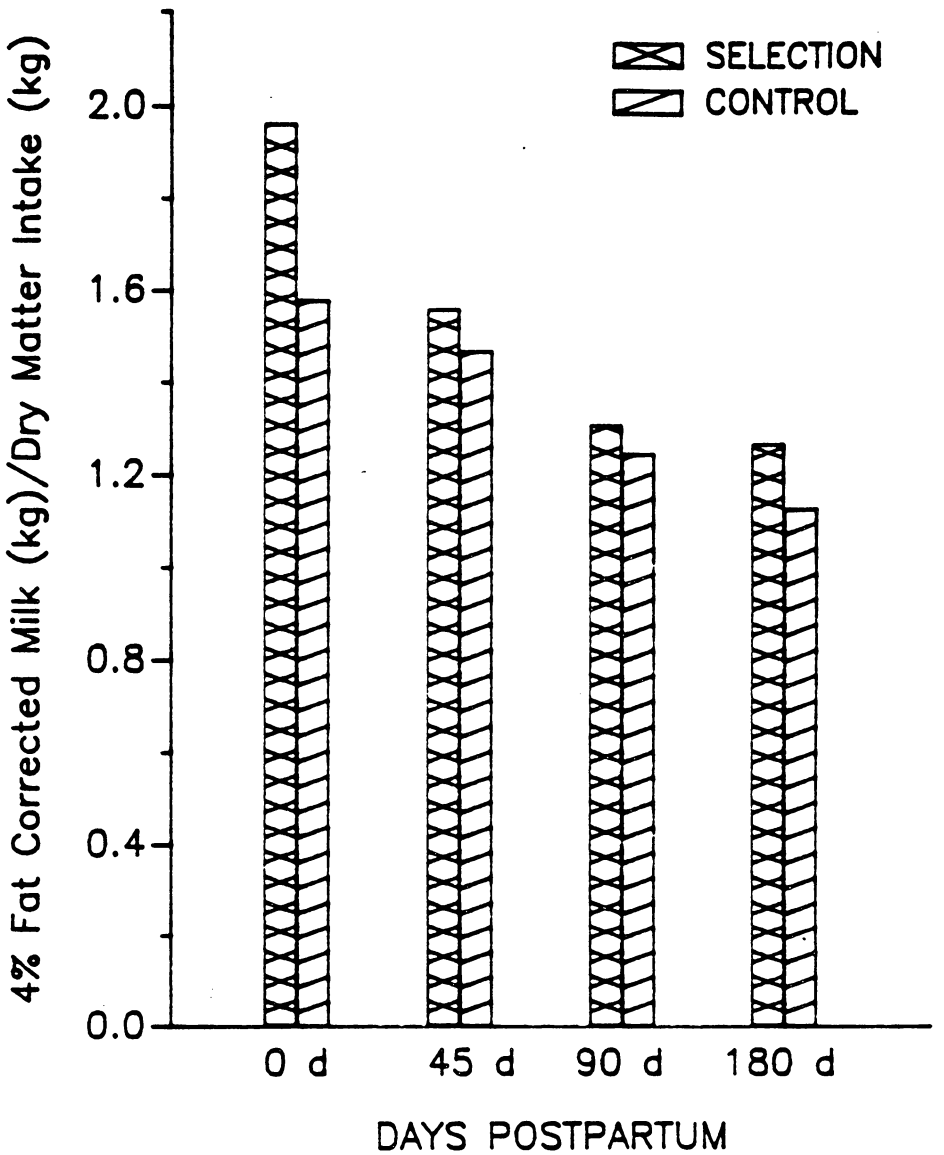

Figure 3. Ratio of 4% fat corrected milk (FCM) to dry matter intake (DMI) in selection and control Holstein cows 0, 45, 90 and 180 days postpartum (dpp), mean standard error is .07

MBW in DM and then decreased to lowest values during the dry period when selection and control cows were consuming 8.0% of their MBW in DM.

There was no significant difference between MBW of selection and control group animals during the first 44 wk of lactation (Figure 2). However, during the first 6 weeks of lactation, selection cows lost weight more rapidly than control cows, 4% vs <1% of initial MBW, respectively (Table 1). Selection cows reached lowest MBW at 3 wk postpartum while control cows reached their lowest MBW 5 wk postpartum (Figure 2). Selection cows took 12 wk to regain their original calving weight while control cows took 8 wk to regain original calving weight (Figure 2).

Selection cows had a greater ratio of kg FCM/kg DMI ($P<.05$) than control cows (Figure 3). The greatest ratio difference between the 2 groups was at 0 dpp, when ratios were 2.0 ± .07 vs 1.6 ± .07, respectively. As lactation advanced, the difference in the efficiency of DMI energy conversion to kg FCM produced between the selection and control cows decreased to 1.3 ± .07 vs 1.1 ± .07, respectively, at 180 dpp (Figure 3). This represented a 35% and a 31% decrease in the ratio of kg FCM/kg DMI in selection and control group animals, respectively.

At 0 and 45 dpp, selection group animals were in a more negative energy balance than control cows, while at 90

and 180 dpp, selection cows were in a less positive NEB ($P<.05$) than control cows (Table 2). As lactation advanced, NEB became less negative ($P<.01$) in both groups (Table 2). Selection and control cows were in a negative NEB at parturition and 45 dpp and a positive NEB at 90 and 180 dpp. During the early dry period, control cows tended ($P<.17$) to be in a more negative energy balance than selection cows.

Plasma glucose concentration at parturition in both groups was greater ($P<.01$) compared to other stages of lactation and the dry period (Figure 4). A selection group by days postpartum interaction ($P<.01$) indicated that at parturition and on the 14 d of the dry period, basal plasma glucose concentration was significantly lower ($P<.01$) in selection cows compared to control cows while concentration at 45, 90 and 180 dpp were similar between genetic groups (Table 3). During the dry period, basal and response period plasma glucose concentrations in selection cows were lower than during lactation, while basal and response period plasma glucose concentrations in control cows during the dry period were greater ($P<.01$) than at 45 dpp and 180 dpp concentrations, (Table 3) (Figure 5, 6). The GRF administered at 0, 45, and 180 dpp and 14 d dry period did not alter plasma glucose in either genetic group (Figure 4).

TABLE 2. Least squares means of net energy balance (NEB) in lactating and dry selection and control cows

Days postpartum[1]	Genetic merit[2]	
	selection	control
0	−8.8±.8[3]	−5.2±.9
45	−3.7±.8	−2.1±.9
90	0.3±.8	1.1±.9
180	0.5±.9	1.6±.9
dry	−2.4±.8	−3.2±1.0

[1]Cows at 0<45<90<180 days postpartum ($p<.0001$)

[2]Lactating selection cows < lactating control cows and dry control cows < dry selection cows ($p<.05$)

[3]Least squares means ± SE, MCAL

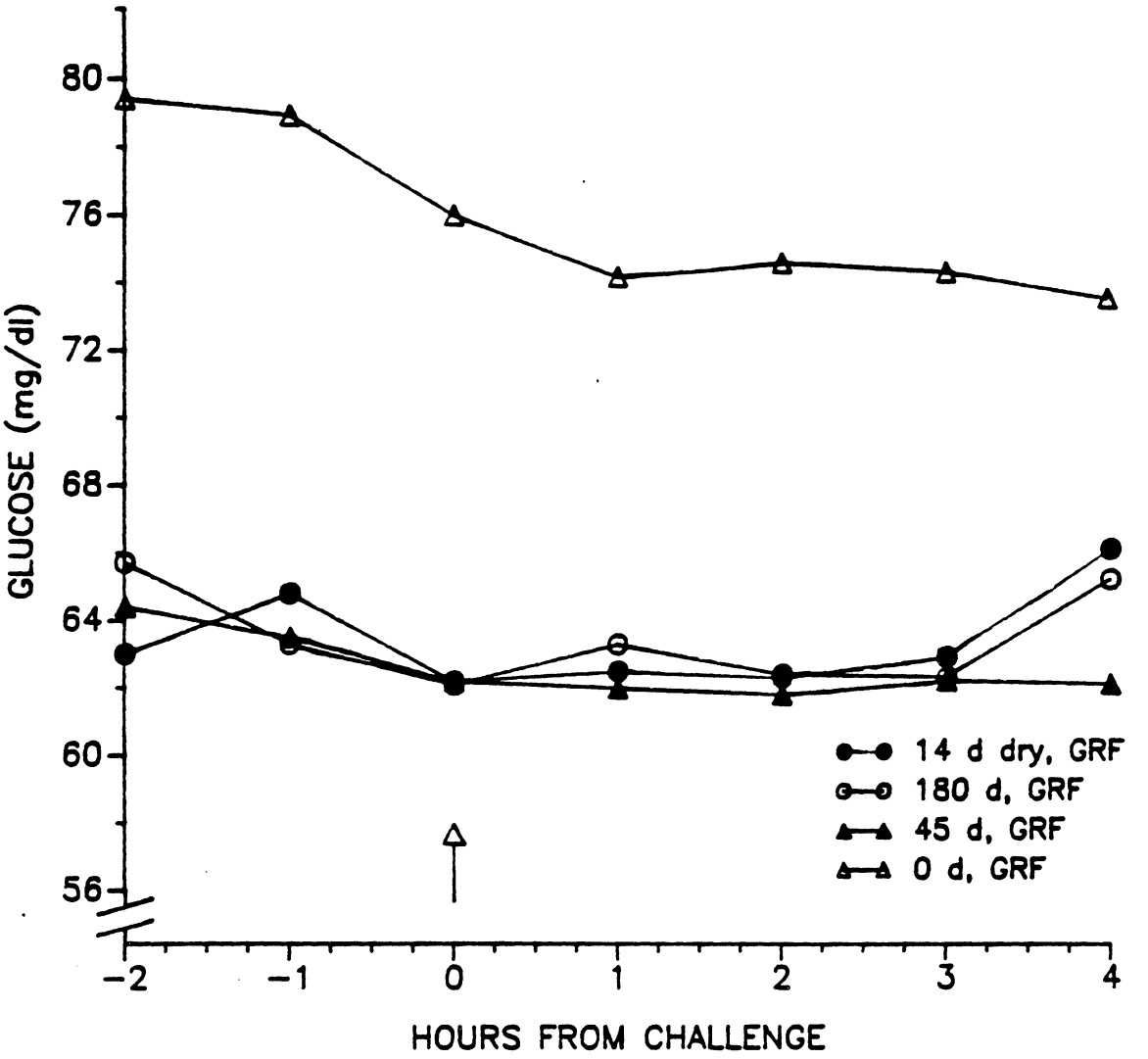

Figure 4. Plasma glucose concentration in primiparous Holstein cows 0, 45, 180 d postpartum and 14 d of the dry period before and after exogenous growth hormone releasing factor (GRF) administration (⍋) (.2 ug/kg body weight), mean standard error is 1.2 mg/dl

TABLE 3. Least squares means of plasma glucose concentration in lactating and dry selection and control cows before and after exogenous growth hormone releasing factor or epinephrine administration[1]

Period[2]	Days[3] postpartum	Genetic merit[4] selection	control
basal	0	75.8± .9	80.4±1.0
	45	64.4± .9	62.4±1.0
	90	65.2± .9	65.0±1.0
	180	63.8± .9	63.6±1.0
	dry[5]	61.3±1.0	65.4±1.1
response	0[6]	71.1± .8	77.2± .9
	45[6]	62.1± .8	62.0± .9
	90[7]	67.9± .8	69.7± .8
	180[6]	62.9± .6	63.7± .8
	dry[6]	60.5± .8	66.4± .9

[1]Least square means ± SE, mg/dl

[2]Days postpartum * period ($p<.01$)

[3]Days postpartum ($p<.01$)

[4]Days postpartum * selection group ($p<.01$)

[5]Day 14 of the dry period

[6]GRF challenge (.2 ug/kg body weight)

[7]Epinephrine challenge (.7 ug/kg body weight)

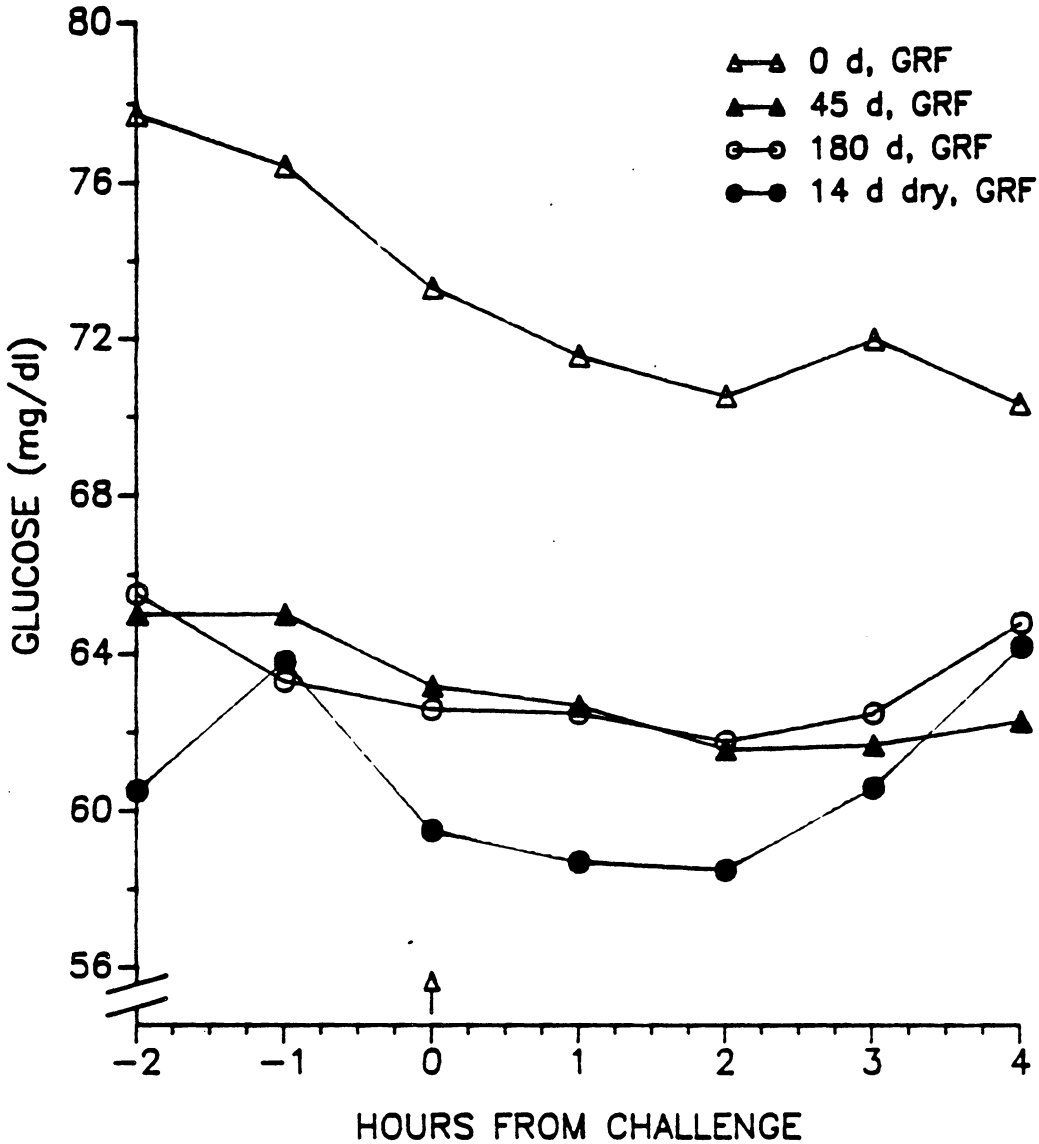

Figure 5. Plasma glucose concentration of selection Holstein cows at 0, 45, 180 d postpartum and 14 d of the dry period before and after growth hormone releasing factor (GRF) administration (Δ) (.2 ug/kg body weight), mean standard error is 1.6 mg/dl

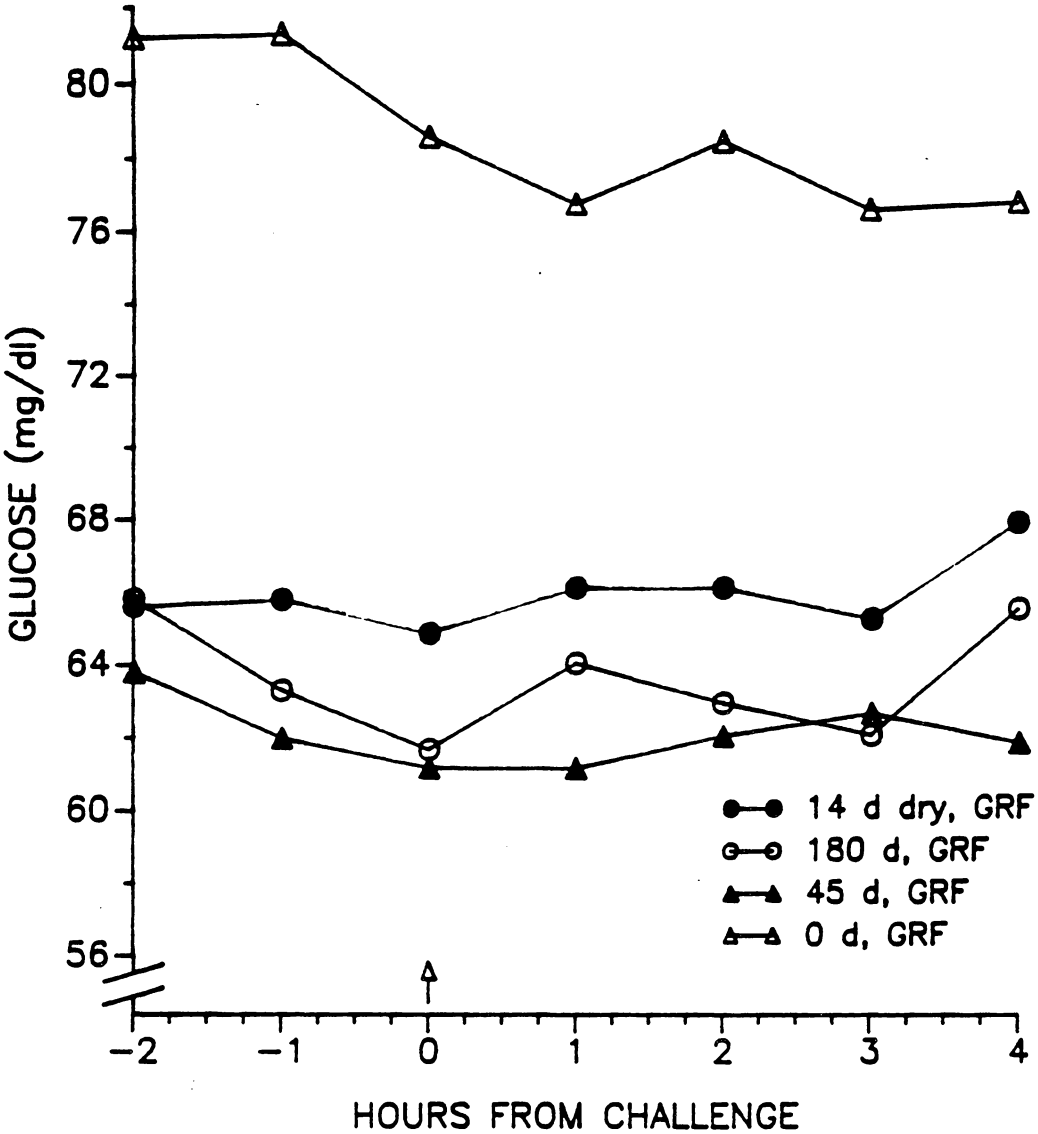

Figure 6. Plasma glucose concentration of control Holstein cows at 0, 45, 180 d postpartum and 14 d of the dry period before and after growth hormone releasing factor (GRF) administration (↑) (.2 ug/kg body weight), mean standard error is 1.7 mg/dl

Basal plasma GH concentration decreased in both groups with advancing lactation. Basal plasma GH concentration was highest at parturition and lowest during the dry period ($P<.01$) (Figure 7). Overall, GH concentration was greater ($P<.05$) in selection cows than control cows (Table 4). A days postpartum by selection group by period interaction ($P<.01$) indicated that at parturition, basal GH concentration was greater in selection cows than in control cows (Table 4), while GH response to the exogenous GRF challenge and return to baseline concentration in 3 hr was similar between genetic groups (figure 8). At 45 dpp, basal GH concentration was similar between groups, but selection cows experienced a greater response and were slower to return to GH baseline concentration than control cows (Table 4, Figure 9). The days postpartum by selection group by period interaction ($P<.01$) also indicated that at 90 dpp basal GH concentration was greater in selection cows than in control cows (Table 4) while at 180 dpp, selection and control cows had similar basal GH concentrations and returned to baseline concentration after the GRF challenge in the same 3.5 hr post-challenge period. However, selection cows ($P<.01$) had a greater GH response to the GRF challenge than control cows at 180 dpp (Table 4, Figure 10). During the dry period, basal GH concentration, return to baseline concentrations and response to the GRF

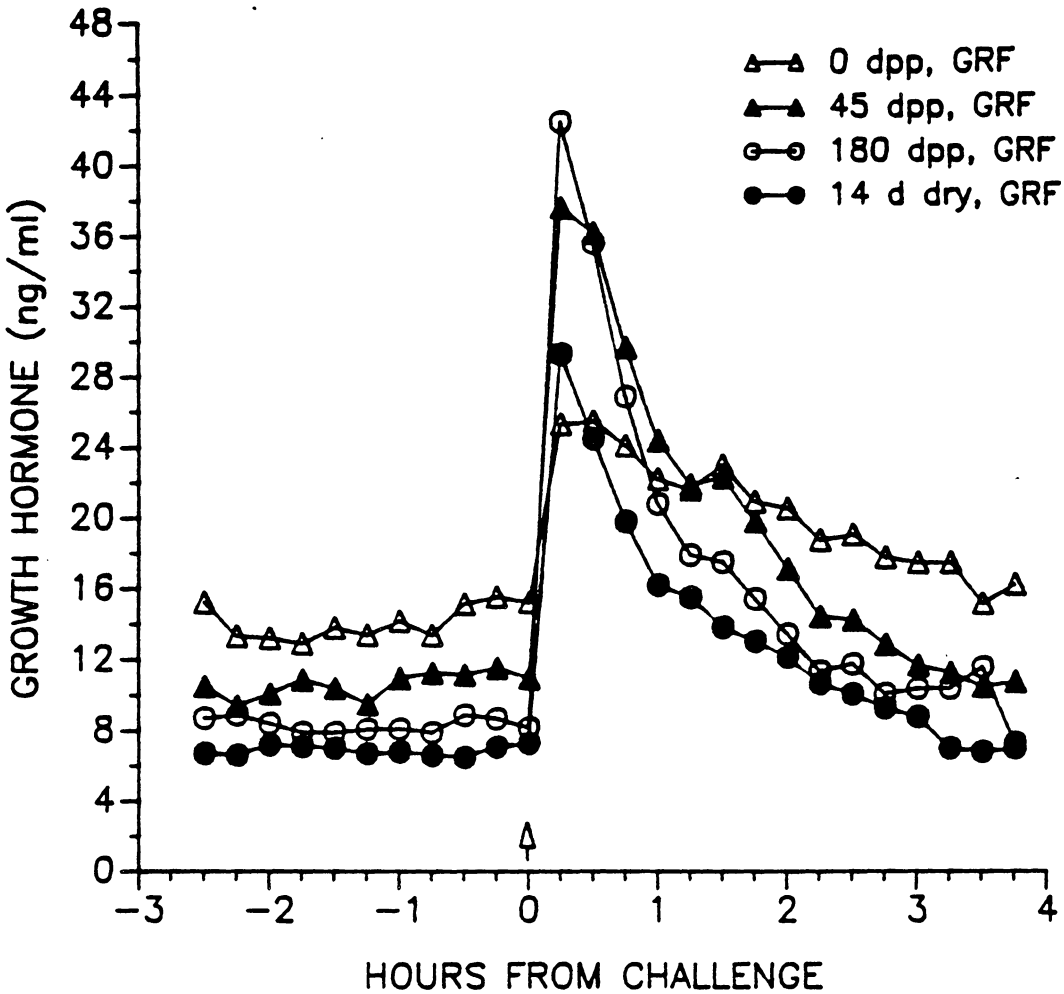

Figure 7. Plasma growth hormone concentration of primiparous Holstein cows at 0, 45, 180 d postpartum and 14 d of the dry period before and after exogenous growth hormone releasing factor administration (Δ) (.2 ug/kg body weight), mean standard error is 1.1 ng/ml

TABLE 4. Least squares means of plasma growth hormone concentration in lactating and dry selection and control cows before and after exogenous growth hormone releasing factor or epinephrine administration[1]

Period[2]	Days[3] postpartum	Genetic merit[4] selection	control
basal	0	15.3±.5	13.0±.5
	45	11.0±.5	10.2±.5
	90	12.3±.4	8.3±.5
	180	9.2±.5	7.5±.5
	dry[5]	7.5±.5	6.3±.5
response	0[6]	19.5±.4	21.3±.4
	45[6]	21.9±.4	17.5±.4
	90[7]	12.5±.4	8.6±.4
	180[6]	18.6±.4	15.5±.4
	dry[6]	13.3±.4	14.0±.5

[1]Least square means ± SE, ng/ml

[2]Basal period < response period ($p<.01$)

[3]Days postpartum ($p<.01$)

[4]Selection cows > control cows ($p<.05$)

[5]Day 14 of the dry period

[6]GRF challenge (.2 ug/kg body weight)

[7]Epinephrine challenge (.7 ug/kg body weight)

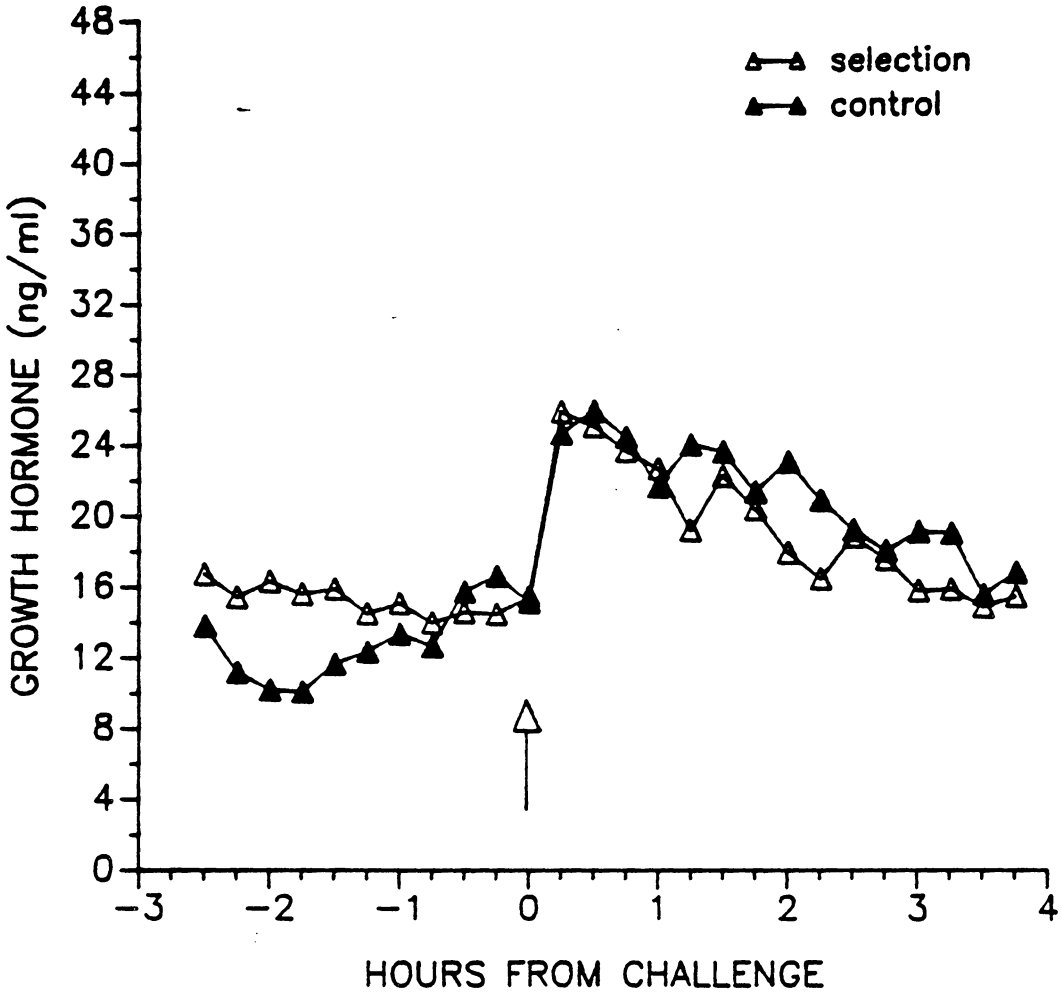

Figure 8. Plasma growth hormone concentration in lactating selection and control Holstein cows at parturition before and after exogenous growth hormone releasing factor administration (Δ) (.2 ug/kg body weight), mean standard error is 1.1 ng/ml

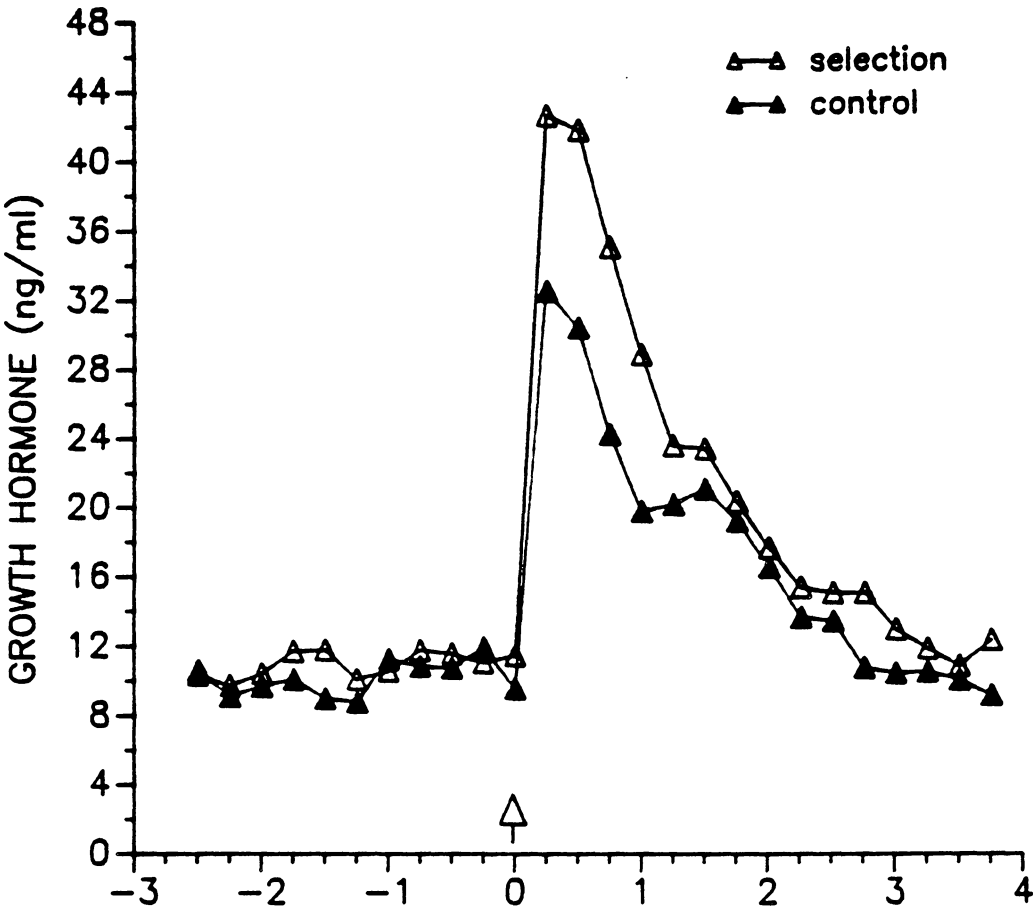

Figure 9. Plasma growth hormone concentration in lactating selection and control Holstein cows at 45 d postpartum before and after exogenous growth hormone releasing factor administration (Δ) (.2 ug/kg body weight), mean standard error is 1.1 ng/ml

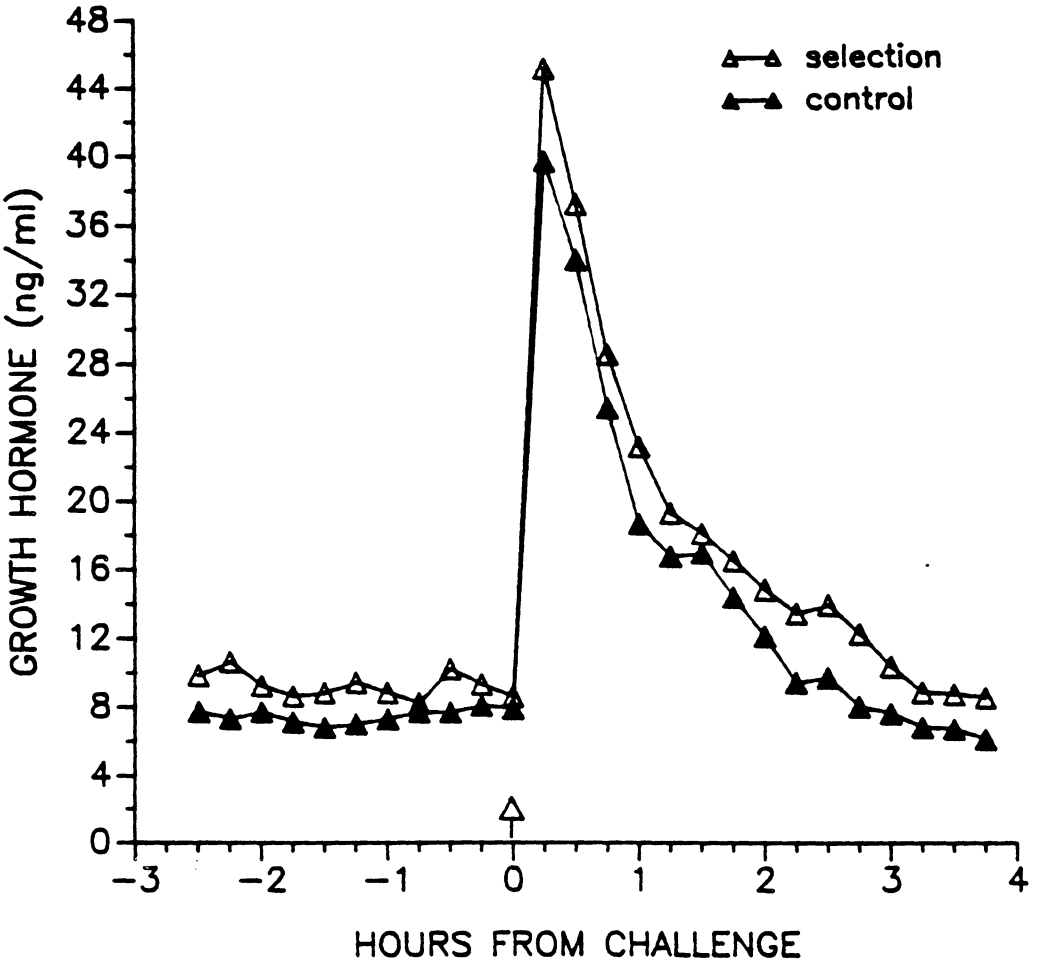

Figure 10. Plasma growth hormone concentration in lactating selection and control Holstein cows at 180 d postpartum before and after exogenous growth hormone releasing factor administration (Δ) (.2 ug/kg body weight), mean standard error is 1.1 ng/ml

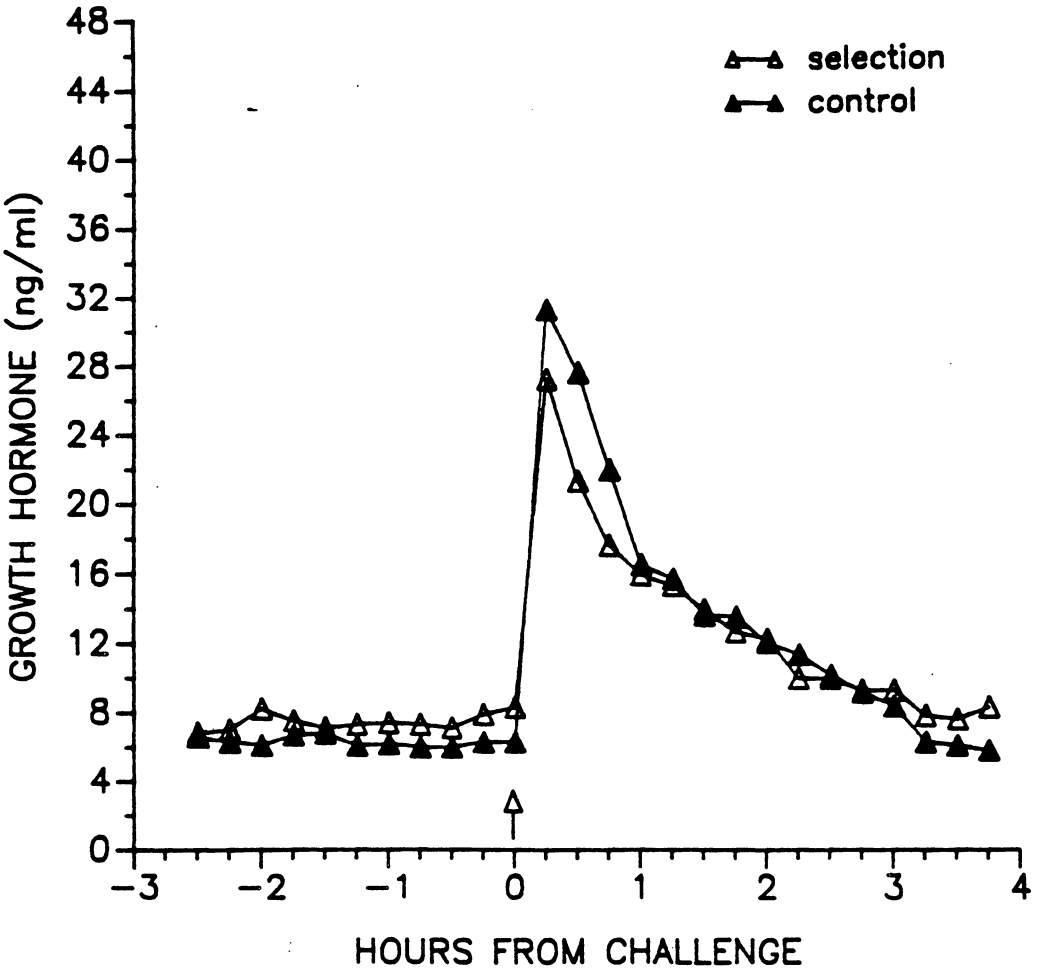

Figure 11. Plasma growth hormone concentration in lactating selection and control Holstein cows at 14 d of the dry period before and after exogenous growth hormone releasing factor administration (↑) (.2 ug/kg body weight), mean standard error is 1.2 ng/ml

challenge were similar between genetic groups (Table 4, Figure 11).

Growth hormone releasing factor stimulated an increase ($P<.01$) in plasma GH concentration in both selection and control cows (Figure 7). A days postpartum by period interaction ($P<.01$) indicated that the GH response to the GRF challenge was greater in later lactation, with the greatest response seen at 180 dpp and the lowest response seen at parturition. The GH response to the GRF challenge at parturition and during the dry period were similar (Figure 7).

A days postpartum by selection group interaction ($P<.01$) indicated that INS concentration of control cows, pre- and post-GRF challenge was lowest at 45 dpp and then increased with advancing lactation, with greatest plasma INS concentration measured on the 14 d of the dry period (Table 5). Similar to control cows, the INS concentration of selection cows also reached lowest values at 45 dpp, however, in contrast to control cows, INS concentration increased to 180 dpp and then leveled off with no further increase during the dry period. Exogenous GRF administration did not stimulate an INS response at 0, 45, 180 dpp or the 14 d of the dry period in either genetic group (Table 5).

TABLE 5. Least squares means of plasma Insulin concentration in lactating and dry selection and control cows before and after exogenous growth hormone releasing factor or epinephrine administration[1]

Period[2]	Days[3] postpartum	Genetic merit	
		selection	control
basal	0	1.2±.03	1.1±.03
	45	1.0±.03	1.0±.03
	90	1.0±.03	1.1±.03
	180	1.2±.03	1.3±.03
	dry[4]	1.2±.03	1.5±.03
response	0[5]	1.1±.03	1.1±.03
	45[5]	1.0±.02	0.9±.03
	90[6]	1.1±.02	1.2±.03
	180[5]	1.2±.03	1.3±.03
	dry[5]	1.3±.03	1.7±.03

[1]Least square means ± SE, ng/ml

[2]Days postpartum * period ($p<.01$)

[3]Days postpartum ($p<.01$)

[4]Day 14 of the dry period

[5]GRF challenge (.2 ug/kg body weight)

[6]Epinephrine challenge (.7 ug/kg body weight)

A days postpartum by selection group interaction ($P<.01$) indicated that while insulin concentrations were similar between the two genetic groups at 0, 45, 90 and 180 dpp (table 5), during the dry period, plasma INS concentration in control cows was greater than in selection cows both before and after infusion of GRF (Figure 12).

Ambient temperature was included in the statistical model as a covariate. Plasma PRL concentration increased with warm temperatures and decreased with cold temperatures ($p<.01$) in both genetic groups. Plasma PRL concentration in both genetic merit groups was the greatest at parturition and the lowest during the early dry period ($P<.01$) (Table 6, Figure 13). A selection group by days postpartum interaction ($p<.01$) indicated that basal and response period PRL concentrations were greater in selection than in control cows on the day of parturition (Figure 14) (Table 6) while plasma PRL concentrations were greater in control than in selection cows at 45 and 90 dpp (Figure 15, 16) (Table 6). Plasma PRL concentrations were similar between genetic groups at 180 dpp and greater in selection cows than in control cows during the dry period (Figure 17, 18, respectively). Exogenous GRF administered on 0, 45, 180 dpp and on the 14 d of the dry period did not alter plasma prolactin concentration (Figure 13). At 45, 90 and 180 dpp, selection and control cows experienced an

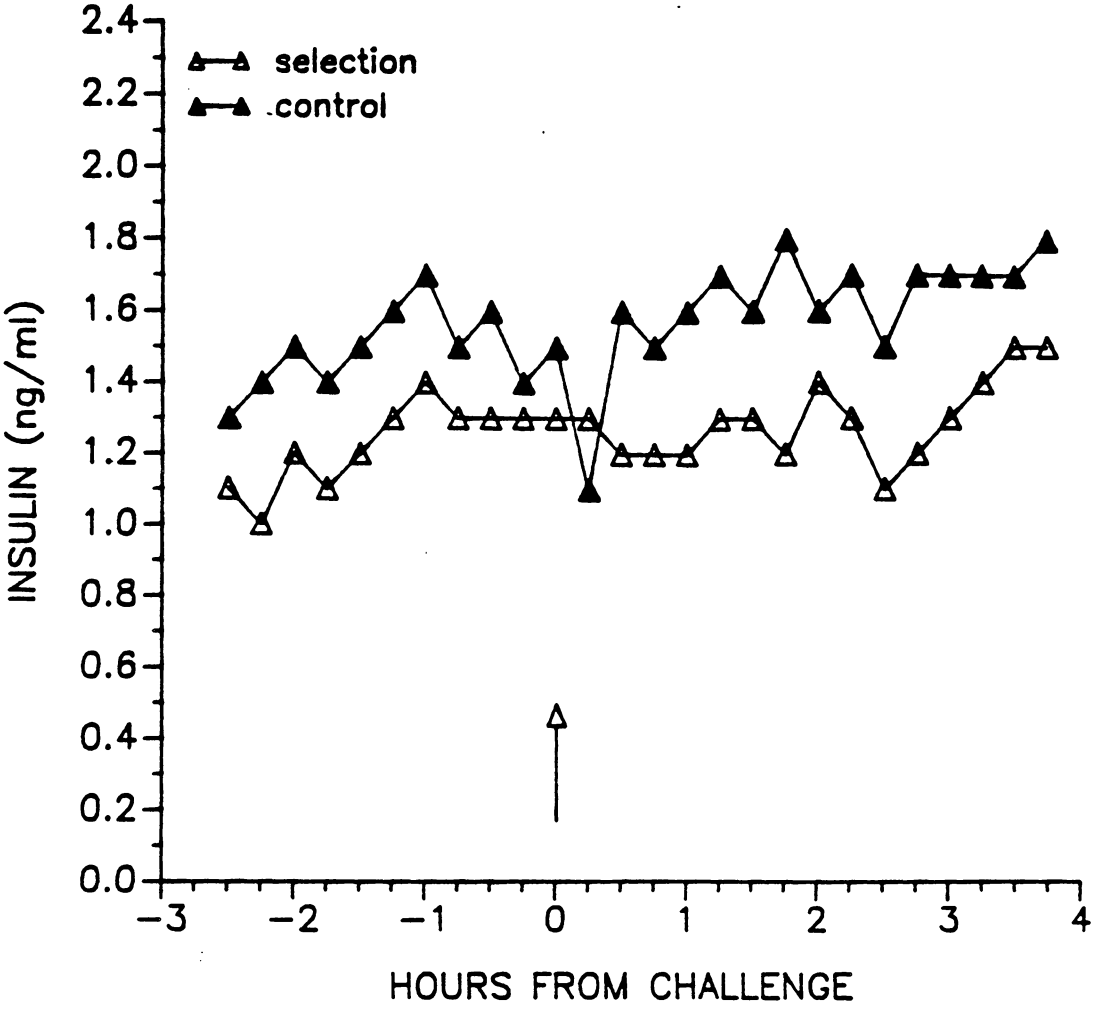

Figure 12. Plasma insulin concentration in selection and control Holstein cows on 14 d of the dry period before and after exogenous growth hormone releasing factor administration (⇧) (.2 ug/kg body weight), mean standard error is .07 ng/ml

TABLE 6. Least squares means of plasma Prolactin concentration in lactating and dry selection and control cows before and after exogenous growth hormone releasing factor or epinephrine administration[1]

Period	Days[2] postpartum	Genetic merit[3] selection	control
basal	0	34.1±.7	30.1±.8
	45	11.8±.7	16.7±.7
	90	12.4±.7	16.4±.8
	180	14.4±.7	14.6±.9
	dry[4]	9.5±.7	4.9±.8
response	0[5]	32.0±.6	28.3±.7
	45[5]	14.1±.6	15.8±.6
	90[6]	10.7±.6	13.7±.7
	180[5]	14.7±.6	15.5±.7
	dry[5]	10.1±.6	7.4±.7

[1]Least square means ± SE, ng/ml

[2]Days postpartum ($p<.01$)

[3]Days postpartum * selection group ($p<.01$)

[4]Day 14 of the dry period

[5]GRF challenge (.2 ug/kg body weight)

[6]Epinephrine challenge (.7 ug/kg body weight)

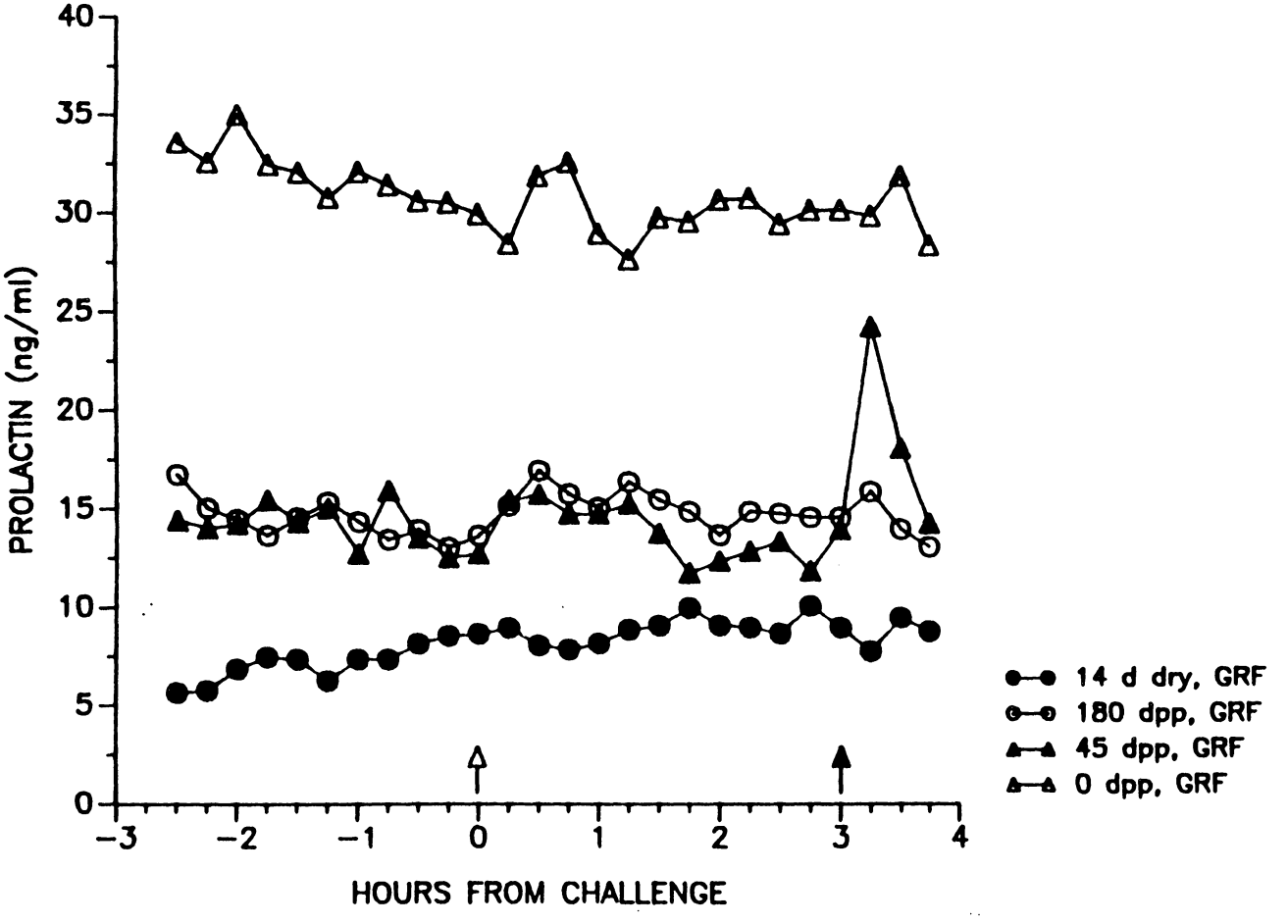

Figure 13. Plasma prolactin concentration in primiparous Holstein cows at 0, 45, 180 d postpartum and 14 d of the dry period before and after exogenous growth hormone releasing factor administration (Δ) (.2 ug/kg body weight). (↑) indicates when animals (45, 180 d postpartum) were milked, mean standard error is 1.7 ng/ml

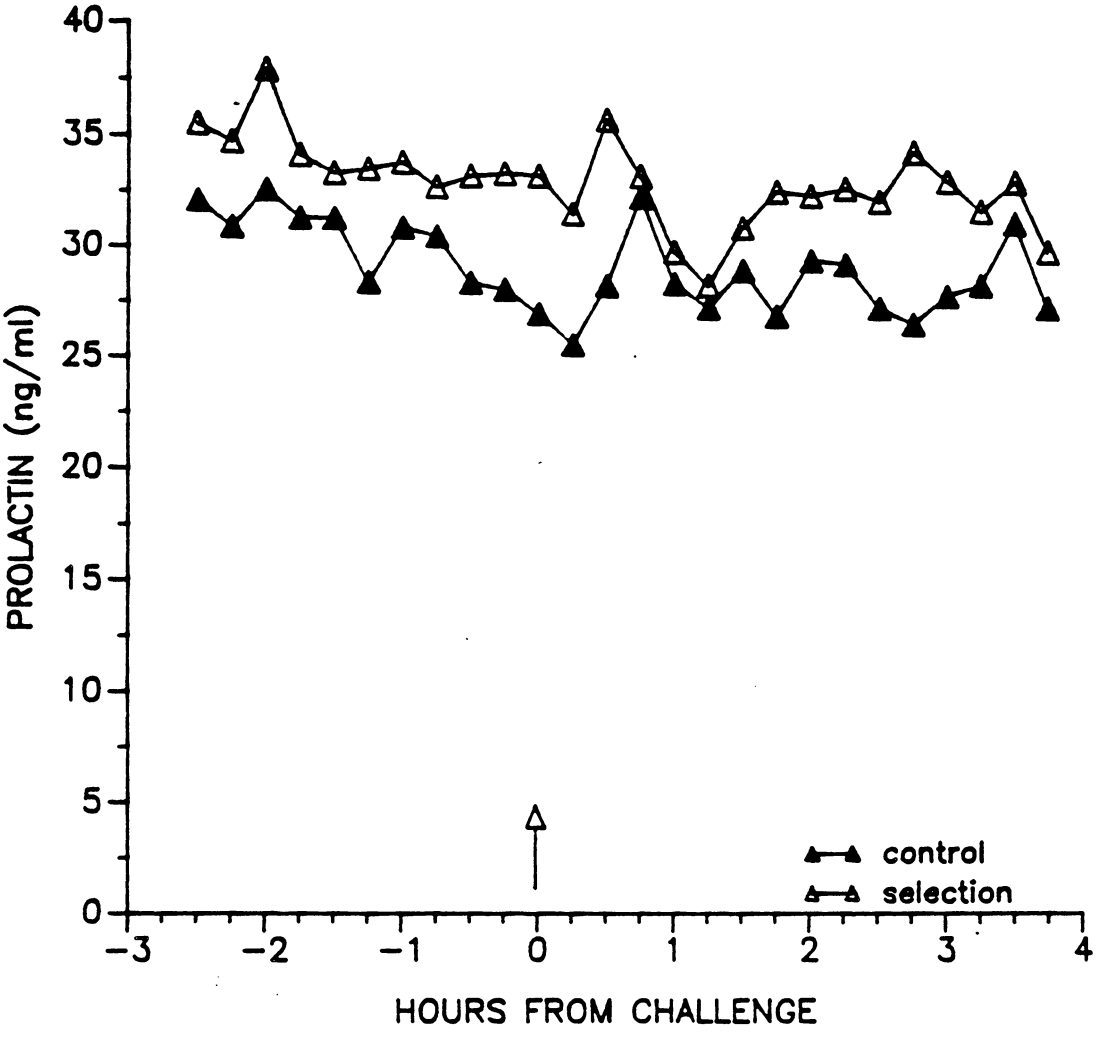

Figure 14. Plasma prolactin concentration in selection and control Holstein cows at parturition before and after exogenous growth hormone releasing factor administration (↑) (.2 ug/kg body weight), mean standard error is 1.7 ng/ml

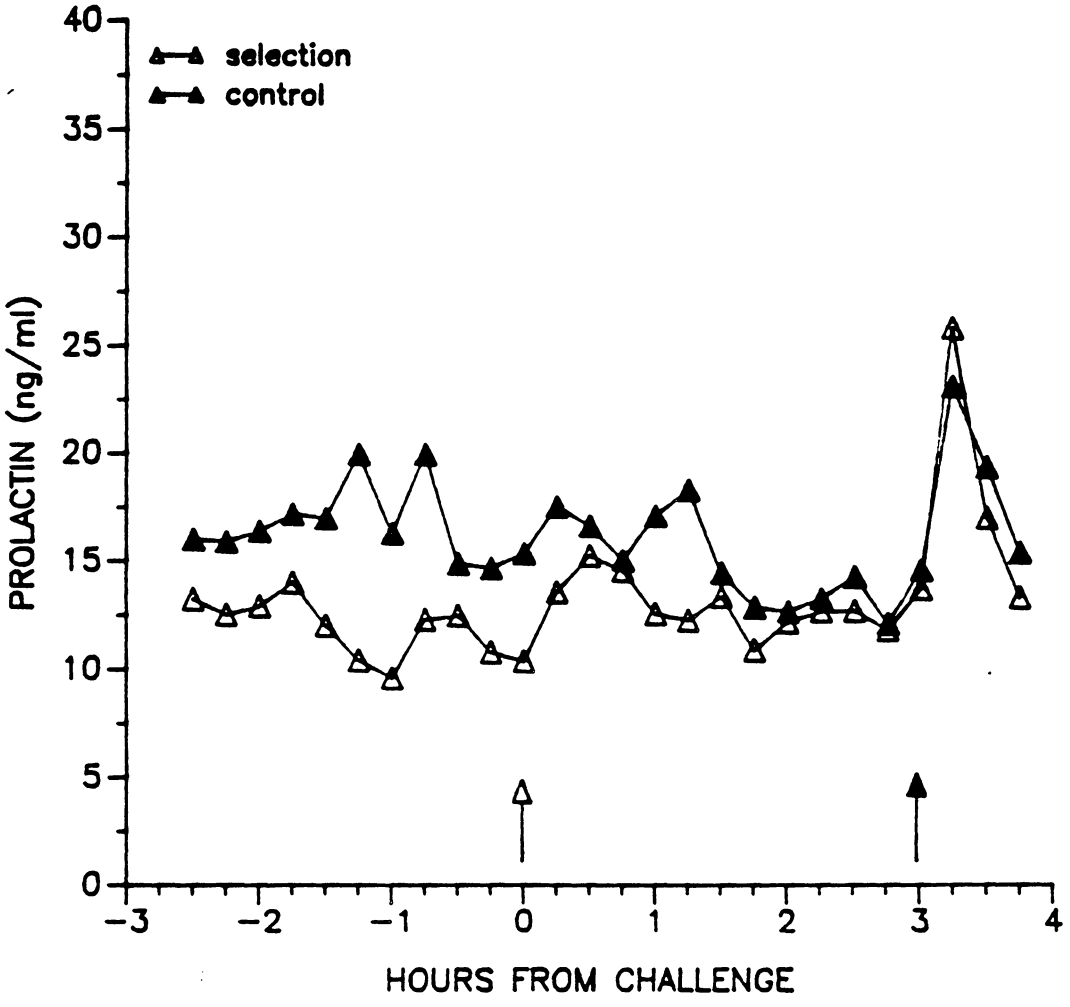

Figure 15. Plasma prolactin concentration in selection and control Holstein cows at 45 d postpartum before and after exogenous growth hormone releasing factor administration (⇧) (.2 ug/kg body weight). (⇩) indicates when animals were milked, mean standard error is 1.7 ng/ml

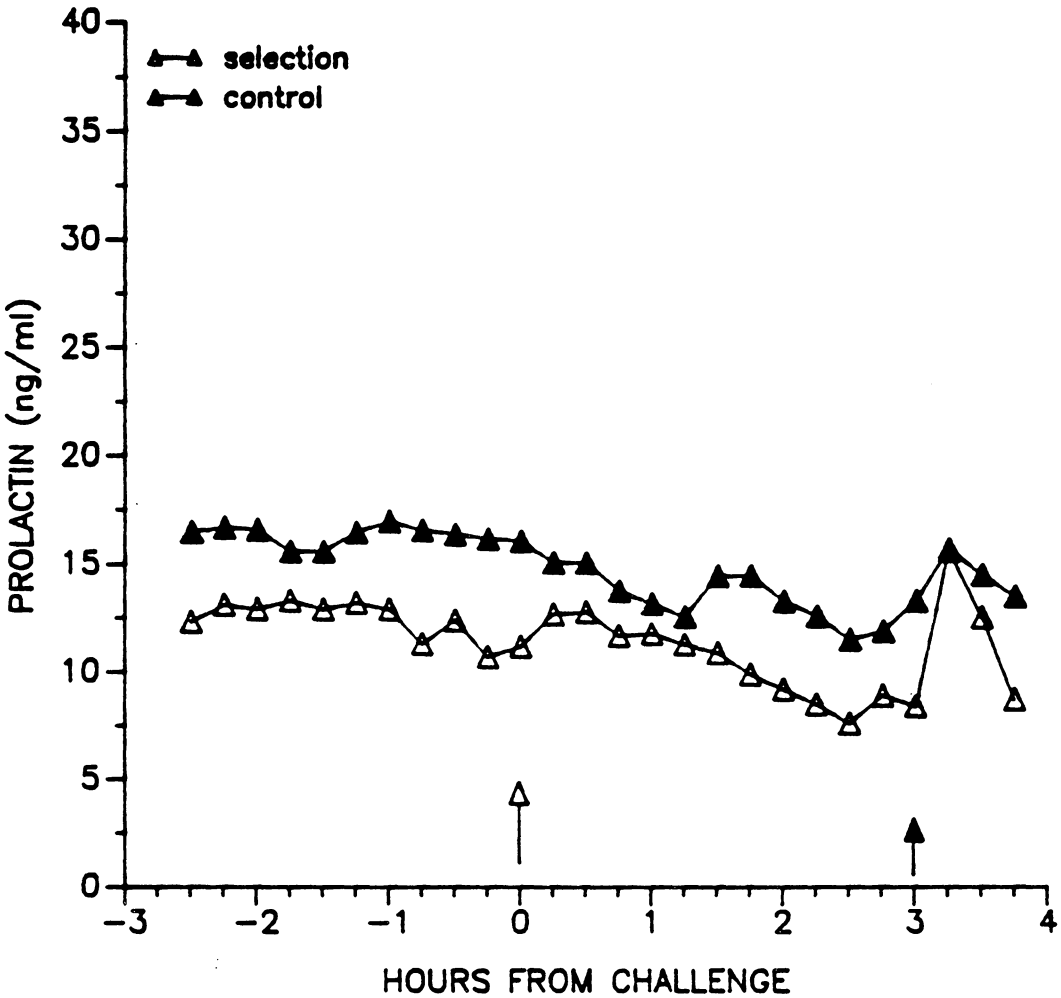

Figure 16. Plasma prolactin concentration in selection and control Holstein cows at 90 d postpartum before and after exogenous epinephrine administration (⍋) (.7 ug/kg body weight), (▲) indicates when animals were milked, mean standard error is 1.7 ng/ml

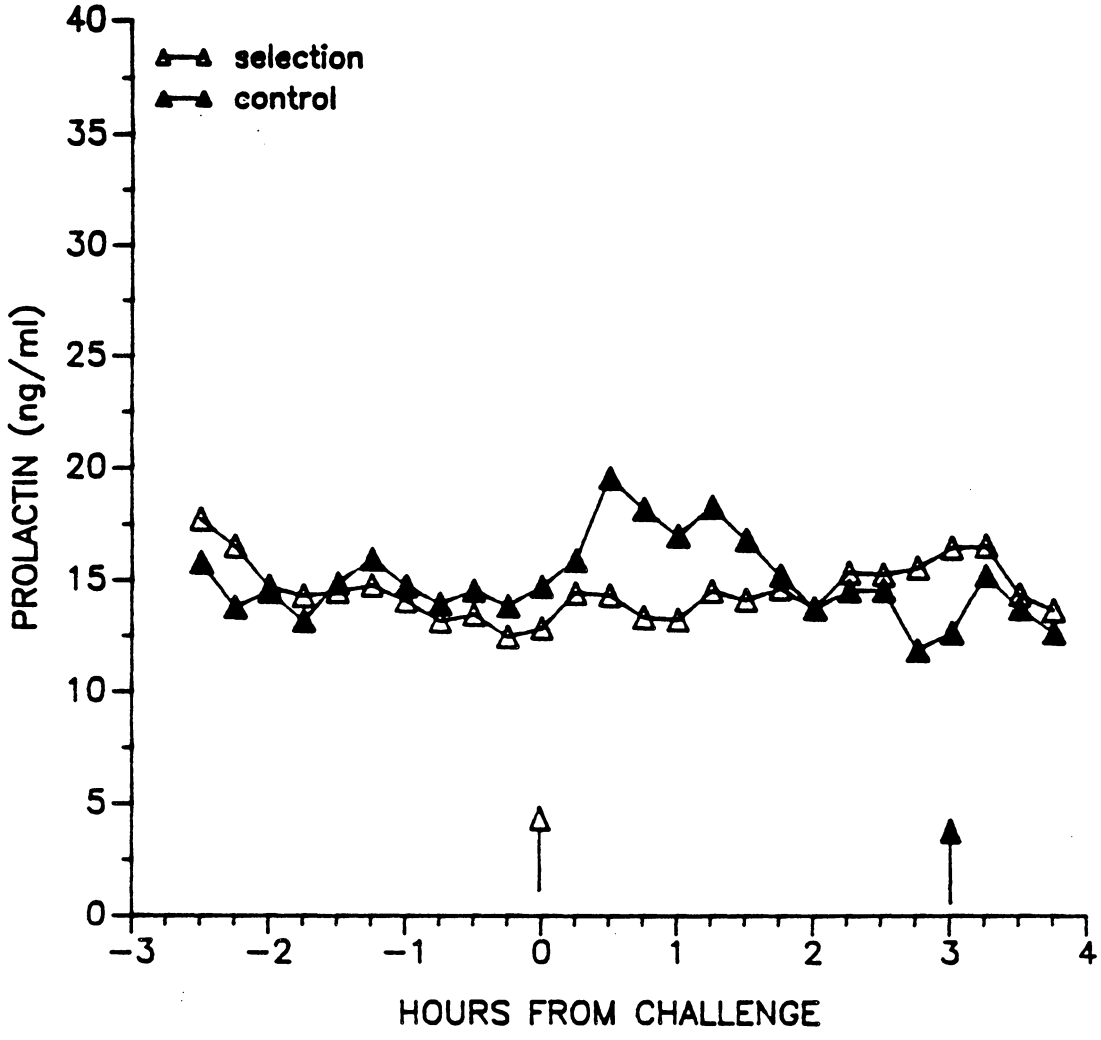

Figure 17. Plasma prolactin concentration in selection and control Holstein cows at 180 d postpartum before and after exogenous growth hormone releasing factor administration (⍙) (.2 ug/kg body weight), (⍙) indicates when animals were milked, mean standard error is 1.7 ng/ml

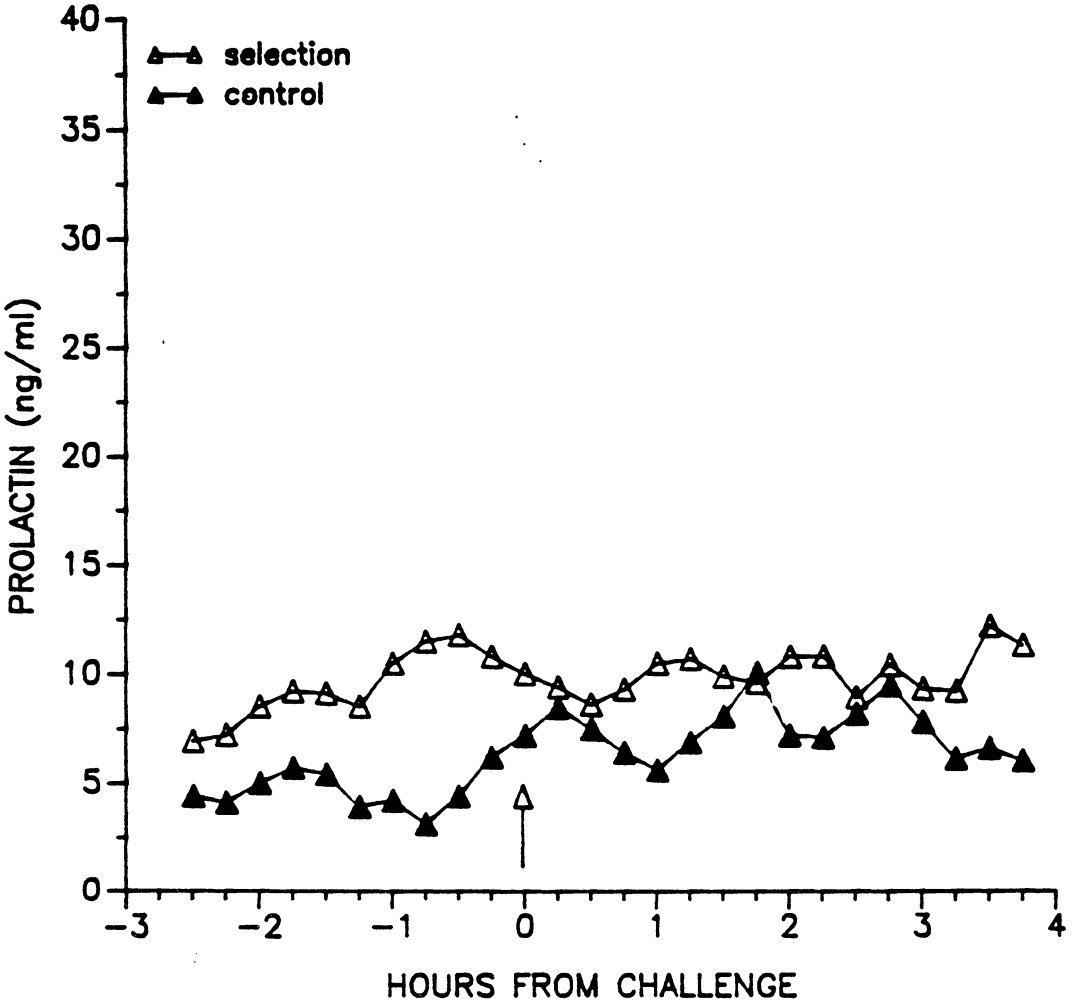

Figure 18. Plasma prolactin concentration in selection and control Holstein cows at 14 d of the dry period before and after exogenous growth hormone releasing factor administration (↑) (.2 ug/kg body weight), mean standard error is 1.8 ng/ml

increase in plasma PRL concentration of varying magnitudes after milking. After the milking stimulus in both genetic groups, at 45, 90 and 180 dpp, plasma PRL concentrations increased by 43%, 31% and 8% respectively, above the plasma PRL concentration in the blood sample taken immediately before milking. Plasma PRL concentration in both groups returned to baseline concentration within an hr of milking (Figure 15, 16, 17, respectively).

Basal and response period plasma concentrations of NEFA was similar between selection and control cows at 90 dpp (Table 7). Concentrations of NEFA differed ($p<.01$) between periods and showed a biphasic increase after epinephrine administration (Figure 19). Epinephrine stimulated an increase in blood NEFA in both groups that returned to baseline approximately 30 min post injection. One hr post injection, a continual increase in plasma NEFA concentration occurred in both groups (figure 19 and table 7).

Exogenous epinephrine administered at 90 dpp stimulated an increase ($P<.01$) in blood glucose concentration in both genetic groups (Figure 20). A days postpartum by period interaction ($P<.01$) indicated that an INS response was stimulated by epinephrine administration in both genetic groups (Figure 21), while exogenous GRF administration at 0, 45, 180 and on the 14 d of the dry

TABLE 7. Plasma concentration of nonesterified fatty acids (NEFA) (uEq/l) in selection and control Holstein cows at 90 days postpartum, before and after exogenous epinephrine[1]

Period [2]	Sample[3]	Genetic merit	
		Selection	Control
basal	1	169.5 ± 9.6[4]	154.9 ± 10.7
	2	159.0 ± 9.6	165.0 ± 10.7
response	3	238.0 ± 9.9	246.3 ± 10.7
	4	152.1 ± 9.9	169.4 ± 10.7
	5	166.8 ± 9.6	131.1 ± 10.7
	6	159.4 ± 9.6	150.5 ± 10.7
	7	177.2 ± 9.6	178.3 ± 10.7
	8	217.3 ± 9.6	227.7 ± 10.7

[1] (.7 ug/kg body weight)

[2] Period 2 > Period 1 ($p<.0002$)

[3] Pooled samples

[4] Least Square Means ± SE, uEq/l

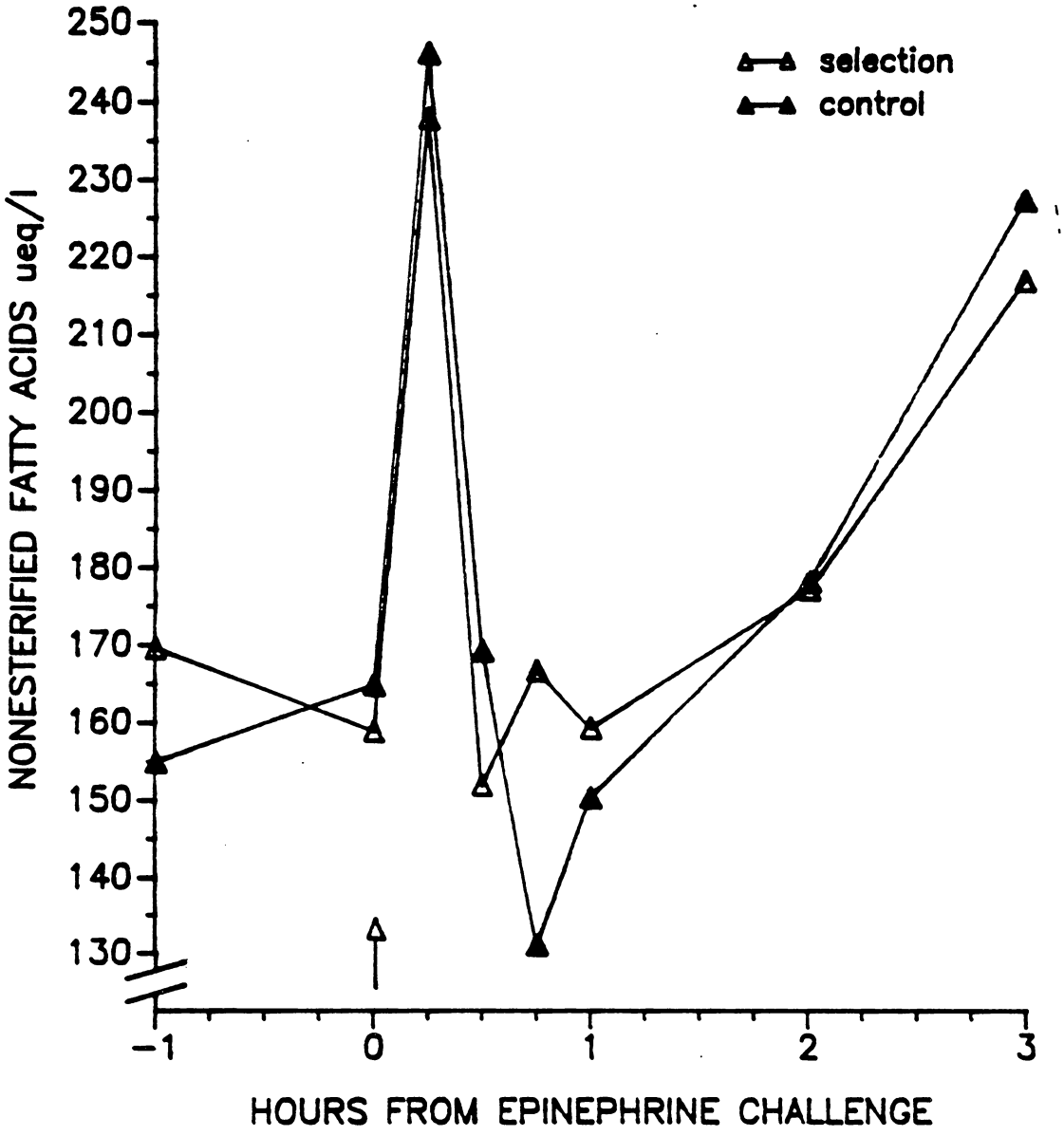

Figure 19. Plasma nonesterified fatty acid concentration (pooled values) in selection and control Holstein cows at 90 d postpartum before and after exogenous epinephrine administration (↑) (.7 ug/kg body weight), mean standard error is 17.0 ueq/l

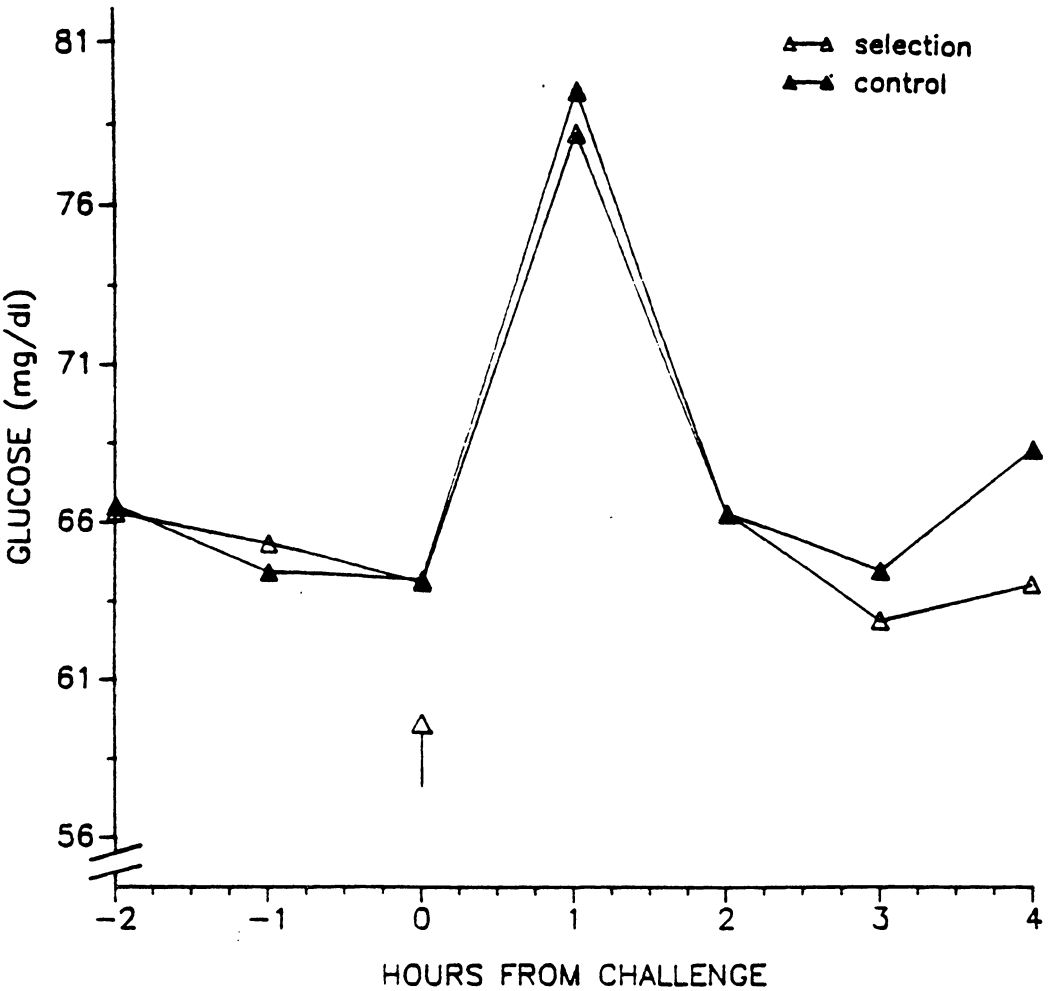

Figure 20. Plasma glucose concentration of selection and control Holstein cows at 90 day postpartum before and after exogenous epinephrine administration (Δ) (.7 ug/kg body weight), mean standard error is 1.7 mg/dl

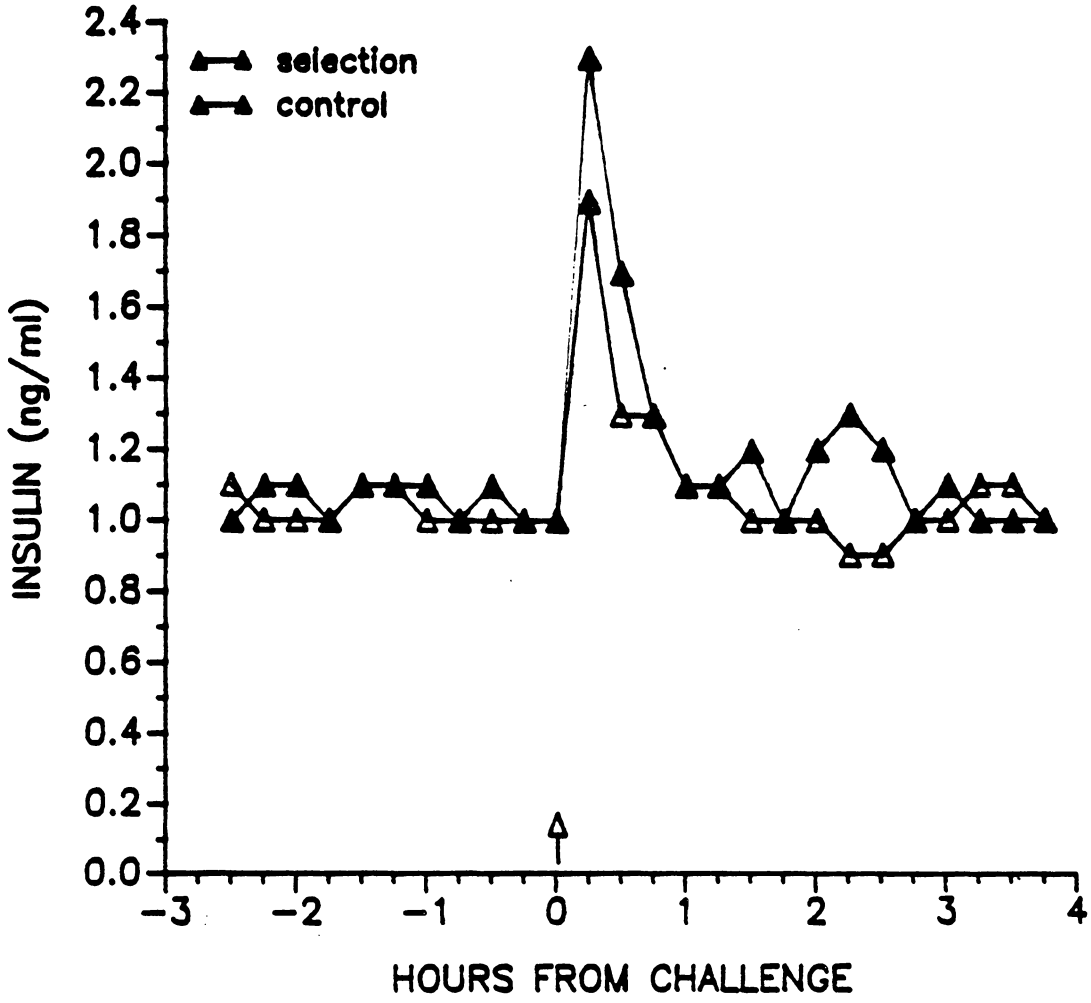

Figure 21. Plasma insulin concentration in selection and control Holstein cows at 90 d postpartum before and after exogenous epinephrine administration (Δ) (.7 ug/kg body weight), mean standard error is .07 ng/ml

period did not stimulate an INS response (Table 5). Exogenous epinephrine administered at 90 dpp did not stimulate a PRL (Figure 16) or a GH (Figure 22) response in selection or control group animals.

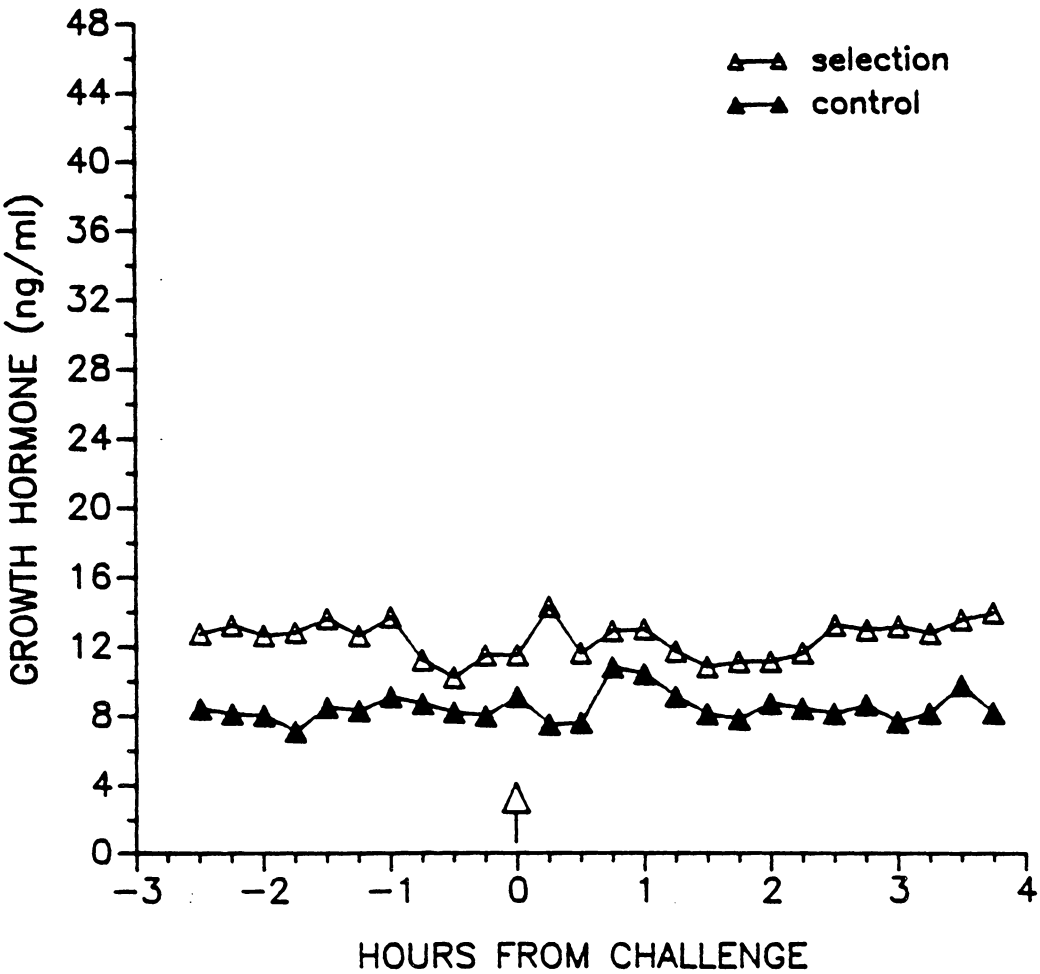

Figure 22. Plasma growth hormone concentration in lactating selection and control Holstein cows at 90 d postpartum before and after exogenous epinephrine administration (Δ) (.7 ug/kg body weight), mean standard error is 1.1 ng/ml

Chapter V

DISCUSSION

Dry matter intake between selection cows and control cows was similar, in contrast to findings of Barnes et al. (1985) and Kazmer et al. (1986) that DMI was significantly greater in first lactation daughters of A. I. sires, compared to lower producing control cows. In agreement with the present study, McNamara and Hillers (1986) reported no difference in DMI between high-yielding and low-yielding Holsteins in a study conducted to investigate possible differences in lipolysis and lipogenesis in cultured adipose tissue from cows of differing genetic merit for milk production. Miller et al. (1987) reported that selection for milk yield can be practiced without causing large increases in feed consumption. In the present study, DMI, at the dpp measured, peaked at 90 dpp and then decreased into the early dry period. Barnes et al. (1987) and Kazmer et al. (1986) recorded peak DMI in primiparous Holsteins between 30 and 90 dpp, when energy was in high demand due to peak milk production, followed by a leveling off and then decline in DMI between 90 and 200 dpp when the energy demand declined with decreasing milk production.

Selection cows were characterized in the present study as more efficient producers at 0, 45, 90 and 180 dpp

because selection cows consumed similar DMI, while having greater milk yields compared to control cows, especially in early lactation. Bryant and Trigg (1981) found cows of higher genetic merit for milk yield were more efficient producers in terms of utilizing feed intake for milk production compared to cows of lesser genetic merit for milk yield.

Hart et al. (1979) investigated differences in endocrine control of energy metabolism between-high yielding Holsteins, and low-yielding Holstein-Hereford crossbreeds. In Hart's study, higher-producing cattle lost weight to wk 14 of lactation and did not regain original calving weight until wk 30 of lactation, while the lower-producing cattle did not lose weight in early lactation but gained weight continually throughout lactation. In Hart's experiment, high and low yielding cows were fed the same quantity of an identical ration. Therefore, the higher-yielding Holstein cows were being underfed with respect to energy needs for production and rapidly lost weight, while lower-yielding control cows were being overfed with respect to production needs and gained weight. However, when high- and low-yielding dairy cattle are fed ad libitum, as in the present study, and in other studies (Bines et al. 1983; Barnes et al.,1985; Kazmer et al.,1986) the pattern of weight loss in early lactation and gradual increase in BW

during later lactation with declining demand for energy for milk synthesis is well documented. These studies have also demonstrated that control cows regain calving weight more quickly than selection cows during lactation.

Selection cows were in a more negative NEB at 0 and 45 dpp and a less positive NEB at 90 and 180 dpp compared to control cows. While selection and control cows had similar DMI, the high-producing group yielded 632 kg more 4% FCM than lower-producing control cows, thus creating a greater energy deficit in selection than in control cows. The fact that selection cows regained their original calving weight more slowly during lactation compared to control cows was also a result of their being in a more negative NEB.

Although the calculated value for NEB is only an estimate of energy inputs and losses, it provides a necessary framework of reference for comparing energy status between groups of differing genetic merit. Both Moe (1981) and Bauman et al. (1980) reported that genetically superior dairy cows tended to prioritize nutrients to be used for milk synthesis and therefore were in a more negative NEB than lower-yielding cows, especially in early lactation. In contrast to these findings, Kazmer et al. (1986) reported that first and second lactation selection cows, genetically selected for high milk yield, and control cows, respectively, and fed a total mixed ration ad

libitum, had a similar calculated NEB throughout lactation. Kazmer et al. (1986) reported a negative NEB at 30 dpp and a positive NEB at 90 and 200 dpp in both selection and control cows.

In the present study selection and control cows were in a negative NEB at parturition and 45 dpp, corresponding to the period of post-calving weight loss and increased milk yield in both groups. However, selection cows were in a more negative NEB than control cows as reflected by their greater weight loss and elevated milk yield compared to control cows during early lactation. Both selection and control cows were in a positive NEB at 90 and 180 dpp corresponding to the period of continual weight gain and decreasing milk yield. Control cows were in a more positive NEB than selection cows at 90 and 180 dpp, and since DMI was similar between genetic groups, the increased energy in control cows may have resulted from the decreased energy used for milk synthesis compared to selection cows. During the early dry period, both selection and control cows were in a negative NEB, although they were consuming a dry cow ration ad libitum and not expending energy on milk production. However, a problem with the palatability of the low energy ration was evident, and because cows weren't allowed to graze or have free choice of hay during the data collection period, it is possible that dry cows from both

genetic groups weren't consuming enough energy from their diet to meet their increased maintenance requirements for pregnancy.

Athanasiou et al. (1978) found that plasma glucose concentration began to increase 2 d pre-partum and peaked at 70.8 mg/dl on the day of parturition in multiparous Holsteins. In the present study, the mean basal glucose concentrations in selection and control group animals at parturition were in agreement with the range reported by Athanasiou and co-workers (1978). Plasma glucose concentration at parturition was greater in selection and control cows compared to concentrations at 45, 90, 180 dpp and 14 d of the dry period. Glucose availability to the mammary gland for lactose synthesis is one of the primary factors limiting milk production (Kronfeld, 1976) and glucose is also required by the mammary gland for synthesis of glycerol of milk lipids and for the supply of the reducing equivalents NADPH for lipogenesis (Bennink et al., 1972). Therefore, plasma glucose concentration may reflect the ability of dairy cattle to synthesize certain milk components as well as absolute milk yield.

In both selection and control cows, plasma glucose concentration tended to decrease from 0 to 45 dpp, increase from 45 to 90 dpp and then decline to 180 dpp. This is in contrast to findings of Herbein et al. (1985) that glucose

concentration continually increased during the first 7 mo of lactation, then stabilized and decreased only in the last 2 mo of lactation in Holsteins of mixed age and genetic merit. The decrease in blood glucose concentration at 180 dpp in the present study may have resulted from a combination of lower DMI and lower body reserves characteristic of first calf heifers in comparison to the more mature cows used in Herbein's study.

A days postpartum by selection group interaction indicated that at parturition and 14 d of the dry period, higher-yielding selection cows had lower plasma glucose concentration than lower-yielding control cows. Lactation and pregnancy have been shown to increase glucose requirements in ruminants, (Armstrong, 1965) therefore, it is possible that during the early postpartum period when selection cows are producing more efficiently than control cows and losing weight more rapidly, selection cows had a larger glucose drain compared to control cows. Alternatively, decreased glucose concentrations in selection cows at parturition and during the early dry period compared to control cows, may be due to differences in the regulation of glucose partitioning by the actions of GH. Growth hormone stimulates the diversion of glucose away from peripheral tissue deposition (Raben, 1973; Trenkle, 1981), thus making it an available energy source

or substrate for lactose synthesis in the mammary gland. Plasma GH concentration was greater in selection group cows compared to control group cows in early lactation and the early dry period, therefore selection cows may have decreased glucose concentrations due to increased glucose partitioning to the mammary gland as a result of increased GH concentration in the selection group. In the present study there were no differences in glucose concentration between selection and control cows at 90 or 180 dpp, similar to findings of Barnes et al. (1985) that blood glucose concentrations in mid-lactation high- and low-yielding Holsteins were similar.

In the present study, in both genetic groups, elevated GH concentration occurred in early lactation in conjunction with greater lactational yield and then gradually decreased in later lactation, corresponding to declining milk yield, until the lowest mean GH concentrations were measured during the early dry period.

Of the hormones measured in the present study, the most consistent difference between genetic groups was in GH concentration. Plasma growth hormone concentration was greater overall in selection compared to control group cows. Kazmer et al. (1986) reported that dairy cows selected on genetic potential for milk yield had greater plasma GH concentrations than control cows before and after

exogenous thyrotropin releasing hormone administration. In the present study, mean basal concentration of selection group animals was greater at 0 and 90 dpp. This is in agreement with findings of Bonczeck et al. (1986) that basal GH concentration was increased in selection cows at peak but not mid-lactation compared to control cows.

On the 14 d of the dry period, basal GH concentrations were similar between cows of differing genetic merit for milk yield. Since plasma GH was not different at all stages measured, this suggests that elevated GH in selection cows during early lactation may have an indirect effect possibly mediated by the increased metabolic demand of lactation and subsequent negative NEB in selection cows and not a direct effect of GH on milk yield. However, a negative NEB would be indicative of this increased metabolic demand, and on the 14 d of the dry period, control cows were in a more negative NEB than selection cows. Therefore, if decreased NEB is associated with elevated GH concentration then elevated GH concentration in control cows would be expected, when in fact, on the 14 d of the dry period, basal GH concentration in control and selection cows was similar. Therefore, the role of GH in selection for increased yield is still unclear.

In the present study, GRF stimulated a GH response in both selection and control groups, at every stage of

lactation measured. However, the magnitude of the response varied with stage of lactation; with greater GRF induced GH response occurring with advancing lactation. Sartin et al. (1985) reported that the GH response to GRF challenge increased in magnitude with advancing lactation due to decreasing concentrations of somatostatin, a small peptide secreted by the central nervous system that inhibits GH release. Elevated endogenous GH concentration has been reported to stimulate hypothalamic somatostatin release (Berelowitz et al., 1981). In the present study, plasma GH concentrations were elevated in early lactation and decreased with advancing lactation. Therefore somatostatin concentration may have been increased in early lactation due to the elevated GH concentration and decreased in later lactation with declining plasma GH concentrations. This potential decline in plasma somatostatin concentration with advancing lactation corresponds with increased plasma GH response to the GRF administration with advancing lactation. Since GH response to GRF was similar during the early dry period and at parturition in both genetic groups, somatostatin may have already begun to increase from low concentrations during late lactation to higher pre-parturition concentrations. Analysis of somatostatin concentrations throughout lactation in selection and

control group animals is needed to further characterize the relationship between somatostatin and GH in the ruminant.

Enright et al. (1986) took continuous samples after administering GRF in Holstein steers and reported peak GH within approximately 20 min post-injection. Similarly, in the present study, peak plasma GH was measured 15 min post-injection. However it is possible that peak GH response occurred before the 15 min post injection sample, and additional, more frequent sampling would be required to ascertain actual peak hormone concentration.

The increased GH response to exogenous GRF in selection cows at 45 and 180 dpp compared to control cows may be due to an increased intrinsic sensitivity of the anterior pituitary to GRF compared to control cows or possibly greater anterior pituitary GH stores in selection cows. Alternatively, selection cows may have decreased somatostatin concentration or be refractory in early to mid lactation to somatostatin's inhibitory effect. At 14 d of the dry period, plasma GH response to exogenous GRF administration was similar between genetic merit groups.

In the present study, basal plasma INS concentration was the reciprocal of GH concentration throughout lactation. Insulin concentration was low in early lactation and gradually increased with advancing lactation; this corresponds to a shift in metabolism from a catabolic

to an anabolic state as lactation progresses and energy demand for milk yield decreases. In the ruminant, INS promotes an anabolic state in effects on carbohydrate, lipid and amino acid metabolism. Insulin stimulates glycogen, FFA and triglyceride synthesis as well as protein synthesis (Basset, 1975).

During the early dry period, plasma INS concentration was greater in control group cows than in selection cows both before and after GRF challenge. This is in agreement with findings of Hart et al. (1980) that INS secretion rate was significantly greater in low-yielding Holstein-Hereford crossbreeds compared to high-yielding Holsteins. However, both low- and high-yielding animals had similar metabolic clearance rates, suggesting that elevated INS concentration in lower-yielding cows may be due to an increased rate of secretion.

In the present study, ambient temperature was shown to affect PRL concentrations. This is in agreement with previous reports that PRL concentration increases in cattle with longer days and warmer temperatures and decreases with shorter days and cooler temperatures (Kazmer et al., 1986).

Plasma PRL concentrations in both selection and control group animals were elevated at parturition compared to 45, 90, 180 dpp and early dry period. Elevated PRL concentrations in periparturient cows have been linked to

lactogenesis (Hartman et al. 1973). Lactogenesis represents a series of events in mammary gland differentiation in which nonsecretory mammary cells are converted into a secretory state. More specifically, this process has been defined as a two-stage mechanism (Fleet et al. 1975, Hartman et al 1973), with the first step consisting of cytologic and enzymatic differentiation of the alveolar cells concurring with limited pre-partum milk secretion. The second stage consists of copious secretion of all products of milk beginning approximately 0 to 4 d pre-parturition and extending through a few days postpartum (Tucker 1981). Schams et al. (1972) reported that blocking the PRL surge just prior to parturition with an ergot alkaloid drug drastically reduced subsequent lactation in cattle, thereby supporting the concept that PRL plays an essential role in the second stage of lactogenesis. Akers (1981) reported PRL to be essential for full lactogenesis in cattle because the absence of PRL during lactogenesis was associated with decreased metabolic activity of mammary cells that were not fully differentiated. In addition, PRL stimulates casein synthesis by increasing casein mRNA transcription (Tucker, 1981).

A days postpartum by selection group interaction indicated that plasma PRL concentration in selection cows

was greater at parturition and during the early dry period compared to control cows. The elevated PRL concentration in selection cows at parturition may induce differentiation of a larger number of alveolar cells capable of synthesizing milk than in early lactating control cows or alternatively, elevated plasma PRL concentration may stimulate PRL receptor numbers of the mammary gland, thus amplifying the effects of PRL on alveolar cells. Although elevated plasma PRL concentration plays an essential role in lactogenesis at parturition (Akers et al., 1981), it is not rate limiting for maintenance of lactation, since suppression of endogenous PRL secretion during an established lactation does not affect milk yield (Smith et al., 1974). Therefore, in addition to the lactogenic function of PRL at parturition, a metabolic function for PRL has been postulated. Forbes (1975) suggested an anabolic role for PRL because he found lambs with increased growth rates had greater PRL concentrations. Therefore, elevated PRL concentration in control cows at 45, 90 and 180 dpp compared to selection cows may be responsible for control cows regaining weight lost post-calving more quickly than selection cows.

On the 14 d of the dry period, plasma PRL concentration in the higher yielding selection group was greater than in the lower yielding control group, agreeing

with findings of Hart et al. (1980) that during the dry period, PRL concentrations increased in higher-yielding Holsteins compared to PRL concentrations of lower-yielding Holstein-Hereford crossbreeds. Hart attributed the increase in PRL concentration to increased PRL secretion rates, possibly in preparation for the lactogenic stimulus at parturition. Elevated PRL at parturition in selection cows compared to control cows in the present study supports this hypothesis.

Selection and control cows exhibited elevated PRL concentrations after milking at 45, 90 and 180 dpp, but the PRL increase due to milking decreased with advancing lactation. Koprowski and Tucker (1973) measured PRL increases from 2.8-fold to as much as 10-fold in some cows due to milking stimulus. These researchers also found a decrease in the PRL response to the milking stimulus with advancing lactation. They hypothesized that the decrease in PRL was due to a decrease in the amount of PRL available for release from the pituitary as lactation advanced. Alternatively, Johke et al. (1970) hypothesized that a gradual reduction in the sensitivity of the neuroendocrine reflex responsible for PRL release occurred in the later stages of lactation.

Basal plasma concentrations of NEFA were similar between selection and control cows at 90 dpp, in contrast

to findings of Bines and Hart (1978) and McNamara and Hillers (1986). Bines and Hart (1978) found concentrations of NEFA were greater in high- than in low-yielding Holsteins up to 120 dpp, and with advancing lactation, this difference decreased until 150 dpp when plasma NEFA concentrations were similar between groups. Barnes et al. (1985) reported that NEFA concentration was elevated in 6 and 24 mo-old selection Holsteins compared to control Holsteins and hypothesized that the NEFA increase in selection animals was due to elevated GH concentrations in these animals compared to controls. McNamara and Hillers (1986) found basal NEFA release from adipose tissue in vitro was greater in high- yielding Holsteins compared to low-yielders, at all stages of lactation.

A possible reason for the similar NEFA concentration between genetic groups in the present study may be related to the small number of samples analyzed in pool form. The addition of a parallel in vitro analysis similar to the study of McNamara and Hillers (1986) would create a more encompassing picture of basal NEFA concentrations between primiparous Holsteins of differing genetic merit for milk production.

Adipose tissue in mammals becomes sensitized to catecholamine stimulus in late pregnancy and lactation (McNamara and Hillers, 1986; Bauman and Currie, 1980).

This sensitization presumably is in preparation for the increased mobilization of adipose tissue for energy at parturition when the cow's metabolism must shift toward support of elevated milk synthesis. Therefore at 90 dpp, cows from both selection and control genetic groups were challenged with epinephrine to determine any potential differences in the intrinsic ability of adipose tissue to adapt to a homeostatic challenge. Both selection and control cows exhibited a similar NEFA release in response to the epinephrine, paralleling the findings of McNamara and Hillers (1986). These researchers reported that although basal NEFA release from cultured adipose tissue was greater in cows selected for high milk yield compared to lower producing control cows, similar adipose tissue NEFA release occurred in both groups in response to the addition of exogenous norepinephrine and epinephrine.

Due to the similarity of basal and response plasma NEFA concentrations between genetic groups in the present study, it can be concluded that genetic selection for milk yield did not alter epinephrine stimulated lipolysis at 90 dpp. However, further experimentation in early and late lactation and during the dry period is needed to characterize the influence of genetic selection on lipolysis throughout the entire reproductive cycle of the dairy cow.

Late lactation, multiparous Holsteins injected with exogenous bGH had a significantly greater NEFA response to exogenous epinephrine administration than control cows receiving no exogenous bGH (McCutcheon and Bauman, 1986). Growth hormone was theorized to be a homeorhetic regulator that influenced nutrient partitioning in cattle by altering the responsiveness of adipose tissue to homeostatic signals. However, in the present study, endogenous GH concentration at 90 dpp was greater in selection group cows compared to control group cows, and the mean basal and response NEFA concentration were similar between genetic groups, regardless. Therefore, differences in physiological concentrations of GH at 90 dpp, apparently did not result in alteration of adipose tissue responsiveness, suggesting that other factors must also play a role in the regulation of NEFA synthesis.

Exogenous epinephrine administration at 90 dpp stimulated glucose release in both selection and control cows, while GRF challenge administered at 0, 45, 180 dpp and on the 14 d of the dry period did not alter glucose. McCutcheon and Bauman (1986) administered daily bGH injections for 12 consecutive days in late lactation multiparous Holsteins, and administered epinephrine on d 13. An increase in plasma glucose of approximately 20 mg/dl resulted from the exogenous epinephrine

administration in the cows that received bGH injections and, although not significant, suggested the potential for GH injections to cause increased glucose production in liver via increased gluconeogenesis and glycogenolysis rates or decreased glucose utilization by body tissues or both. In the present study, epinephrine administration induced a stress-related response in the cows that resulted in increased plasma glucose levels possibly via increased gluconeogensis or glycogenolysis. Control cows tended to have a greater mean glucose response to the epinephrine challenge than selection cows. A possible explanation for this finding may be an increased gluconeogenic rate accompanied with decreased peripheral tissue utilization of glucose and an inefficiency in control cows to effectively partition a large portion of glucose to the mammary gland. Alternatively, because at 90 dpp, control cows had similar DMI and a tendency for reduced milk yield compared to selection cows, more substrate may be available for gluconeogenesis in the control cows than in selection cows.

At 90 dpp, the exogenous epinephrine administration stimulated an INS response in both selection and control group cows. In early lactation metabolism of the dairy cow is altered to support high lactational yield. Increased INS in response to exogenous epinephrine administration represented a homeostatic response indicative of a

momentary metabolic adjustment in an attempt to reach a steady metabolic state. Control cows tended to experience greater INS response to exogenous epinephrine than selection group cows. Control cows may have had greater pancreatic INS stores or the pancreas may have been sensitized due to the increased plasma glucose concentration in control animals compared to the selection cows in response to exogenous epinephrine administration. While differences between genetic lines can be detected in blood levels of INS after feeding, after infusion of metabolites or during fasting (Land et al, 1983; Granzer et al., 1983; Schwab et al., 1984, respectively) there are no reports in the literature of differences in plasma INS response to epinephrine administration in dairy cows differing in genetic merit for milk yield.

Chapter VI

CONCLUSION

Results indicated that selection pressure for milk yield in primiparous Holstein cows resulted in more efficient production (kg FCM/kg DMI) at 0, 45, 90 and 180 dpp, increased milk yield, decreased NEB, greater post-partum weight loss, and elevated concentrations of plasma GH confounded by NEB effects. Consistent effects of selection for milk yield on blood PRL, INS, glucose and NEFA were not evident, indicating no apparent direct effects of selection pressure on regulation of these hormones and metabolites in the alteration of production efficiency. Increased milk yield efficiency in selection cows may result from the interactions of GH with these hormones and metabolites or other growth factors such as somatomedins. Alteration of somatomedin ratios with INS and PRL due to GH stimulated somatomedin release may favor a metabolic state in which lipolysis and gluconeogenesis dominate, resulting in increased milk yield efficiency.

The fact that NEFA response to epinephrine was similar in spite of selection pressure indicates that adipose tissue responsiveness to lipolytic stimulus was similar between genetic groups. Thus, while this study indicates that selection pressure for milk yield resulted in

alteration of endocrine relationships that augment production efficiency in cattle, the question remains as to whether this is a direct effect or an effect mediated by differences in NEB. Therefore, because of the possible confounding influence of NEB on the endocrine status in the ruminant, the present study does not support the hypothesis that blood hormone and metabolite concentrations can practically be used as indicators of genetic potential for milk yield during established lactation.

Chapter VII

SUMMARY

This study investigated potential differences in hormone and metabolite concentrations and possible NEB differences between Holstein cows differing in genetic merit for milk yield. In comparing phenotypic performance, selection and control cows had similar dry matter intake, while selection cows produced more milk and more kg FCM per kg DMI than control cows. Net energy balance was negative in early lactation and positive in later lactation in both genetic groups, while NEB of selection cows was decreased compared to control cows throughout lactation.

The most consistent hormonal difference found between high- and low-yielding Holsteins in the present study was elevated plasma GH concentration in selection cows compared to control cows. In both genetic groups, plasma GH and PRL concentrations were elevated at parturition and decreased with advancing lactation, while plasma INS concentration increased with advancing lactation, reaching mean peak concentration during the early dry period. Plasma GH response to exogenous GRF administration increased in both genetic merit groups with advancing lactation. However, a consistent metabolite difference between high- and low-yielding Holsteins was not apparent. Basal NEFA concentration was similar between genetic groups at 90 dpp,

while blood glucose was elevated at parturition compared to concentrations measured during lactation and the dry period in both genetic groups. Selection and control cows had a similar plasma NEFA and glucose response to exogenous epinephrine administration, while control cows tended to experience greater INS response to the epinephrine administration than selection cow

LITERATURE CITED

1. Aguis, L., A. M. Robinson, J. R. Girard, and D. H. Williamson. 1979. Alterations in the rate of lipogenesis in vivo in maternal liver and adipose tissue on premature weaning of lactating rats. Biochem. J. 180:689.

2. Akers, R. M., D. E. Bauman, A. V. Gupuco, G. T. Goodman, and H. Allen Tucker. 1981. Prolactin regulation of milk secretion and biochemical differentiation of mammary epithelial cells in periparturient cows. Endocrinology 109:23.

3. Athanasiou, N., and R. W. Phillips. 1978. Stability of plasma metabolites and hormones in parturient dairy cows. Am J. Vet. Res. 39:953.

4. Armstrong, D. G. 1965. Carbohydrate metabolism in ruminants and energy supply. Pages 272-288 in Physiology of digestion in the ruminant. R. W. Dougherty, R. S. Allen, W. Burroughs, N. L. Jacobson, and A. D. McGilliard, ed. Butterworths, Inc., Washington, D.C.

5. Baile, C. A., and F. C. Buonomo. 1987. Growth hormone releasing factor effects on pituitary function, growth and lactation. J. Dairy Sci. 70:467.

6. Baile, C. A., M. A. Della-fera, and F. C. Buonomo. 1983. Growth hormone releasing factor injections in sheep. J. Anim. Sci. (Suppl 1):188 (abstr).

7. Barnes, M. A., G. W. Kazmer, R. M. Akers and R. E. Pearson. 1985. Influence of selection for milk yield on endogenous hormones and metabolites in Holstein Heifers and cows. J. Anim. Sci. 60:271

8. Barnes, M. A., 1987. Improving production efficiency in dairy cattle. VPI&SU Extension Presentation. Blacksburg, Va. Cow College.

9. Bassett, J. M. 1975. Dietary and gastro-intestinal control of hormones regulating carbohydrate metabolism in ruminants. Page 383 *in*: Digestion and Metabolism in Ruminants. I. W. McDonald and A. C. I. Warner, ed. Univ. New England Publ. Unit. Amidale, Australia.

10. Bauman, D. E., S. N. McCutcheon, W. D. Steinhour, P. J. Eppard, and S. J. Secher. 1985. Sources of variation

and prospects for improvement of production efficiency in the dairy cow: A review. J. Anim. Sci. 60:583.

11. Bauman, D. E., P. J. Eppard, M. J. Greeter and G. M. Lanza. 1985. Responses of high producing dairy cows to longterm treatment with pituitary somatotropin and recombinant somatotropin. J. Dairy Sci. 68:1352

12. Bauman, D. E. and W. B. Currie. 1980. Partitioning of nutrients during pregnancy and lactation: A review of mechanisms involving homeostasis and homeorhesis. J. Dairy Sci. 63:1514.

13. Bauman, D. E., R. M. Akers, L. T. Chapin, H. A. Tucker, and E. M. Convey. 1979. Effect of level of intake on serum concentrations of prolactin and growth hormone in lactating cows. J. Dairy Sci. 62 (Suppl 1):114 (abstr).

14. Bennink, M. R., R. W. Mellenberger, R. A. Frobish and D. E. Bauman. 1972. Glucose oxidation and entry rate as affected by the initiation of lactation. J. Dairy Sci. 55:712(Abstr).

15. Berelowitz, N., M. Szabo, L. A. Frohman, S. Firestone and L. Chu. 1981. Somatomedin-C mediated growth hormone negative feedback by effects on both hypothalamus and the pituitary. Science 212:1279.

16. Bines, J. A., I. C. Hart and S. V. Morant. 1983. Endocrine control of energy metabolism in the cow: Diurnal variations in the concentrations of hormones and metabolites in the blood plasma of beef and dairy cows. Horm. metabol. Res. 15:330.

17. Bines, J. A. and I. C. Hart. 1982. Metabolic limits to milk production, especially roles of growth hormone and insulin. J. Dairy Sci. 65:1375.

18. Bines, J. A. and I. C. Hart. 1978. Hormonal regulation of energy between milk and body tissue in adult cattle. Proc. Nutr. Soc. 37:281.

19. Bonczek, R. R., C. W. Young, J. E. Wheaton, and K. P. Miller. 1986. Correlated response to selection for high milk yield in dairy cattle: Plasma concentrations of growth hormone, prolactin, insulin and thyroxine at two stages of lactation. Ph.D. Thesis, University of Minnesota, St. Paul, Minn.

20. Botts, R. L., R. W. Hemken and L. S. Bull. 1979. Protein reserves in the lactating cow. J. Dairy. Sci. 62:433.

21. Brown, R. E. 1969. The conversion of nutrients into milk, pg. 23 in The University of Nottingham third nutrition conference for feed manufacturers. I. I. Swan and D. Lewis, ed. J & A Churchill LTD., London, England.

22. Byrant, A. M. and T. I. Trigg. 1981. Progress report on the performance of jersey cows differing in breeding index. Proc. New Zeland Soc. Anim. Prod. 41:39.

23. Collier, R. J., J. P. McNamara, C. R. Wallace and M. H. Dehoff. 1984. A review of endocrine regulation of metabolism during lactation. J. Anim. Sci. 59:498.

24. Convey, E. M. 1974. Serum hormone concentrations in ruminants during mammary growth, lactogenesis and lactation: A review. J. Dairy Sci. 57:905.

25. Cotes, P. M., J. A. Crichton, J. F. Folley, and F. G. Young. 1949. Galactopoietic activity of anterior pituitary growth hormone. Nature 164:992.

26. Custodio, A. A., R. W. Blake, P. F. Dahm, T. C. Cartwright, G. T. Schelling and C. E. Coppock. 1983. Relationships between measures of feed efficiency and transmitting ability for milk of Holstein cows. J. Dairy Sci. 66:1937.

27. Dalton, C., and C. Kowalski. 1967. Automatic colorimetric determination of FFA in biological fluids. Clin. Chem. 13:744.

28. Davey, A. M. F., C. Graninger, D. D. S. Mackenzie, D. S. Flux, G. F. Wilson, I. M. Brookes and C. W. Holmes. 1983. Nutritional and physiological studies of differences between Friesian cows of high and low genetic merit. Proc. New Zeland Soc. Anim. Prod. 43:67.

29. Eiseman, J. H., D. E. Bauman, A. C. Hammond, P. J. Reynolds, H. F. Tyrrell and G. L. Haaland. 1984. The influence of growth hormone administration on the kinetics of plasma nonesterified fatty acids in growing Hereford heifers. Can. J. Anim. Sci. 64(Suppl):308.

30. Enright, W. J., L. T. Chapin, W. M. Moseley, S. A. Zinn, and H. Allen Tucker. 1986. Growth hormone

releasing factor stimulates milk production and sustains growth hormone release in Holstein cows. J. Dairy Sci. 69:344.

31. Eppard, D. J., D. E. Bauman and S. N. McCutcheon. 1985. Effect of dose of growth hormone on lactation of dairy cows. J. Dairy Sci. 68:1109.

32. Eppard, D. J., D. E. Bauman, J. Bitman, D. Wood, R. M. Akers and W. A. House. 1985. Effect of dose of bovine growth hormone on milk composition: alpha lactalbumin, fatty acids and mineral elements. J. Dairy Sci. 68:3047.

33. Falconer, J., J. M. Forbes, J. A. Bines, J. H. B. Roy and I. C. Hart. 1980. Somatomedin-like activity in cattle: The effect of breed, lactation, and time of day. J. Endocrinol. 86:183.

34. Feteris, W. A., 1965. A serum glucose method without protein precipitation. Am. J. Med. Technol. 31:17.

35. Fleet, I. R., J. A. Goode, M. H. Harmon, M. S. Laurie, J. L. Linzell and M. Peaker. 1975. Secretory activity of goat mammary glands during pregnancy and the onset of lactation. J. Physiol. 251:763.

36. Flux, D. S., D. D. S. Mckenzie and G. F. Wilson, 1984. Plasma metabolite and hormone concentration in Friesian cows of differing genetic merit measured at 2 feeding levels. Anim. Prod. 38:377.

37. Folley, S. J., and F. G. Young. 1940. Further experimentation on the continued treatment of lactating cows with anterior pituitary extracts. J. of Endocrinology. 2:226.

38. Forbes, J. M., P. M. Driver, A. A. El-Sharat, T. G. Boaz and C. G. Scones. 1975. The effect of day length and level of feeding on serum prolactin in growing lambs. J. Agric. Sci.(Camb) 64:549.

39. Fronk, J. J., C. J. Peel, D. E. Bauman and R. C. Gorewit. 1983. Comparison of different patterns of exogenous growth hormone administration on milk production in Holstein cows. J. Anim. Sci. 57:699.

40. Gorski, J. 1979. Endocrine factors in genetic improvement of milk production. J. Dairy Sci. 62:814.

41. Granzer, W., R. Hahn, F. Pirchner. 1983. Die Insulinkonzentration Zuchtwert. Zuchtungskinde 55:91

42. Hart, I. C. 1983. Endocrine control of nutrient partitioning in lactating ruminants. Proc. Nutr. Soc. 42:181.

43. Hart, I. C., J. A. Bines and S. V. Morant. 1980. The secretion and metabolic clearance rates of growth hormone, insulin, and prolactin in high and low yielding cattle at 4 stages of lactation. Life Sci. 27:1839.

44. Hart, I. C., J. A. Bines and S. V. Morant. 1979. Endocrine control of energy metabolism in the cow: correlations of hormones and metabolites in high and low yielding cows at 4 stages of lactation. J. Dairy Sci. 62:270.

45. Hart, I. C., J. A. Bines, S. V. Morant, and J. L. Ridley. 1978. Endocrine control of energy metabolism in the cow: Comparisons of the levels of hormones (prolactin, growth hormone, insulin, and thyroxine) and metabolites in the plasma of high and low yielding cattle at various stages of lactation. J. Endocrinol. 77:333.

46. Hart, I. C., J. A. Bines, C. C. Balch and A. T. Cowie. 1975. Hormone and metabolic differences between lactating beef and dairy cattle. Life Sci. 16:1285.

47. Hart, I. C. 1973. Effect of 2-bromo-alpha-ergocryptine on milk yield and level of prolactin and growth hormone in the blood of the goat at milking. J. Endocrinology 57:179.

48. Hartman, P. E. 1973. Changes in the composition and yield of the mammary secretion of cows during the initiation of lactation. J. Endocrinol. 59:231.

49. Herbein, J. H., R. J. Aiello, L. I. Eckler, R. E. Pearson, and R. M. Akers. 1985. Glucagon, insulin, growth hormone, and glucose concentration in blood plasma of lactating dairy cows. J. Dairy Sci. 68:320

50. Hickman, C. G. 1980. Physiological effects of selection for milk yield. *in*: Selection Experiments in Laboratory and Domestic Animals. pages 198-211. A. Robertson, ed. Commonw. Agr. Bur. United Kingdom.

51. Ingalls, W. G., E. M. Convey, and H. D. Hafs. 1973. Bovine serum lutenizing hormone, growth hormone, and prolactin during late pregnancy, parturition and early lactation. Proc. Soc. Exp. Biol. Med. 143:161.

52. Johke, T. 1970. Factors affecting plasma prolactin level in the cow and rat as determined by radioimmunoassay. Endocrinol. Jap. 17:393.

53. Kaprowski, J. A. and H. Allen Tucker. 1973. Serum prolactin during various physiological states and its relationship to milk production in the Bovine. Endocrinol. 91:1480.

54. Kazmer, G. W., M. A. Barnes, R. M. Akers and R. E. Pearson. 1986. Effect of genetic selection for milk yield and increased milking frequency on plasma growth hormone and prolactin concentration in Holstein cows. J. Anim. Sci. 63:1220.

55. Ko, H., and M. E. Royer. 1967. A submicrometer assay for nonpolar acids in plasma and depot fat. Anal. Biochem. 20:205.

56. Kronfeld, D. S. 1976. The potential importance of the proportions of gluconeogenic, lipogenic, and aminogenic nutrients in regard to the health and productivity of dairy cows. Adv. Anim. Physiol. Anim. Nutr. 7:5.

57. Kronfeld, D. S., G. P. Mayer, J. M. Robertson and F. Raggi. 1963. Depression of milk secretion during insulin administration. J. Dairy Sci. 46:559.

58. Land, R. B., W. R. Carr, I. C. Hart, T. J. Osmond, R. Thompson and N. Tilakaratne. 1983. Physiological attributes as possible selection criteria for milk production. 3. Plasma hormone concentrations and metabolite and hormonal responses to changes in energy equilibrium. Anim. Prod. 37:165.

59. Land, R. B. 1981. Physiological criteria and genetic selection. Livest. Prod Sci. 8:203.

60. Massri, Y. G., S. N. Al-Raheem, C. W. Young, and J. E. Wheaton. 1985. Growth hormone response to growth hormone releasing factor in selected and control Holstein heifer calves. J. Dairy Sci. 68:1819.

61. McCutcheon, S. N., and D. E. Bauman. 1986. The effect of pattern of administration of bovine growth hormone

on lactation performance of dairy cows. J. Dairy Sci. 69:39.

62. McCutcheon, S. N. and D. E. Bauman. 1986. Effect of chronic growth hormone treatment on response to epinephrine and thyrotropin releasing hormone in lactating cows. J. Dairy Sci. 69:44.

63. McCutcheon, S. M., D. E. Bauman, W. A. Murphy, W. A. Lance and D. H. Coy. 1984. Effect of human synthetic pGRF on plasma growth hormone concentration in lactating cows. J. Dairy Sci. 67:2881.

64. McNamara, J. P., and J. K. Hillers. 1986. Regulation of Bovine adipose tissue metabolism during lactation 2. Lipid response to milk production and energy intake. J. Dairy Sci. 69:3042.

65. Miller, R. H., R. E. Pearson, E. M. Coffey, and L. A. Fulton. 1987. The relationship between daughters' feed intake and predicted difference for milk sires. J. Anim. Breedg. Genet. 104:295.

66. Moe, P. W., 1981. Energy metabolism of dairy cattle. J. Dairy Sci. 64:1120.

67. Moe, P. W., 1965. Effect of level of intake on the utilization of diets by dairy cows. Ph.D. Thesis, Cornell University, Ithica, N.Y.

68. Moseley, W. M., L. F. Krabill, A. R. Hillerman and R. F. Olsen. 1984. Growth hormone response of steers injected with synthetic human pancreatic growth hormone releasing factor. J. Anim. Sci. 58:432.

69. Peel, C. J. and D. E. Bauman. 1987. Somatotropin and lactation. J. Dairy Sci. 70:474.

70. Peel, C. J. T. J. Fronk, D. E. Bauman and R. C. Gorewit. 1983. Effect of exogenous growth hormone in early and late lactation on lactation performance of dairy cows. J. Dairy Sci. 66:776.

71. Peel, C. J., T. J. Fronk, D. E. Bauman, and R. C. Gorewit. 1982. Lactation response to exogenous growth hormone and abomasal infusion of a glucose-caseinate mixture in high yielding dairy cows. J. Nutr. 112:1770.

72. Peel, C. J., D. E. Bauman, R. C. Gorewit, and C. J. Sniffen. 1981. Effect of exogenous growth hormone on

lactation performance in high yielding cows. J. Nutr. 111:1662.

73. Plouzek, C. A., L. L. Anderson, D. L. Hard, J. R. Molina, and A. Trenkle. 1983. The effect of intravenous injection of growth hormone releasing factor on plasma growth hormone in cattle. J. Anim. Sci. (Suppl 1) 57:203 (Abstr).

74. Raben, M. S. 1973. page 261. In: Method in investigative and diagnostic endocrinology S. A. Benson and R. I. Yalow, ed. London: North Holland.

75. Rabinowtiz, D., J. J. Merimee and J. A. Burgess. 1966. Growth hormone - insulin interaction. Fact and Speculation. Diabetes 15:905.

76. Sartin, J. L., R. J. Kemppainen and K. A. Cummins. 1985. Effects of GRF, somatostatin and glucose on growth hormone secretion in lactating dairy cows. Am. J. Physiol. 248:E108.

77. Schams, D., V. Reinhardt, and H. Karg. 1972. Effects of 2-Br-α-ergokryptine on plasma Prolactin level during parturition and onset of lactation in cows. Experientia 28:692.

78. Schwab, M., F. Pirchner, G. Peeters, W. Granzer, W. Groth and R. Hahn. 1984. Physiological reactions before and after feeding in relation to dairy merit of bulls. 35th Ann. Mtg. EAAP.

79. Sejrsen, K., and P. Lovendahl. 1986. Criteria identifying genetic merit for milk prod. Pages 142-152. *in*: Exploiting New Technologies in Animal Breeding. C. Smith, J. W. B. King and J. C. Mckay, ed. Oxford Science Publications.

80. Smith, V. G., T. W. Beck, E. M. Convey, and H. A. Tucker. 1974. Bovine serum prolactin, growth hormone, cortisol and milk yield after ergocryptine. Neuroendocrinology 15:172.

81. Swan, H. 1976. The physiological interrelationship of reproduction, lactation, and nutrition in the cow. Pages 85-102. *in*: Principles of Cattle Production. H. Swan and W. H. Broster, ed. Butterworths, Kent, England.

82. Trenkle, A. 1981. Endocrine regulation of energy metabolism in ruminants. Fed Proc. 40:2536.

83. Tucker, H. A., 1981. Physiological control of mammary growth, lactogenesis and lactation. J. Dairy Sci. 64:403.

84. Vasilatos, R. and P. J. Wangsness. 1981. Diurnal variation in plasma insulin and growth hormone associated with 2 stages of lactation in high producing dairy cows. Endocrinology. 108:300.

85. Wako NEFa in Vitro Enzymatic Colorimetric Method for the Quantitative Determination of Non-Esterified (or Free) Fatty Acids in Serum. 1986. Wako Pure Chemical Industries, Ltd. 990-75401, Osako, Japan)

86. Williams, W. F., Wisshaar, A. G. and G. E. Lauterbach. 1966. Lactogenic hormones effect on plasma nonesterified fatty acids and blood glucose concentrations. J. Dairy Sci. 49:106.

87. Yang, Y. T. and R. L. Baldwin. 1973. Preparation and metabolism of isolated cells form bovine adipose tissues. J. Dairy Sci. 56:350.

88. Young, J. W. 1977. Gluconeogenesis in cattle: Significance and methodology. J. Dairy Sci 60:1.

The vita has been removed from the scanned document